



LIBRARY
New Delhi

Call No. X-II 540

Acc. No. 14590

Indian Agricultural Research Institute (Pusa)
LIBRARY, NEW DELHI-110012

This book can be issued on or after _____

| Return Date | Return Date |
|-------------|-------------|
| | |

be charged.

2. Books may be renewed at the discretion of the Head Library Services.
3. Dog-earing of pages of a book, marking or writing there in with ink or pencil, tearing or taking out its pages or otherwise damaging it will constitute an injury to a book.
4. Unless a borrower points out the injury at the time of borrowing the book, he shall be required to replace the book or pay its price, if detected at the time of return.

**HELP US TO KEEP THIS BOOK
FRESH AND CLEAN**

THE ANALYST

THE JOURNAL OF THE

Society of Public Analysts and other Analytical Chemists

A MONTHLY JOURNAL DEVOTED TO THE ADVANCEMENT
OF ANALYTICAL CHEMISTRY

INDEX TO

VOL. 65

1940

PUBLISHED FOR THE SOCIETY BY

W. HEFFER & SONS LTD.

4, PETTY CURY, CAMBRIDGE, ENGLAND

1940

THE ANALYST

THE JOURNAL OF THE

Society of Public Analysts and other Analytical Chemists

A MONTHLY JOURNAL DEVOTED TO THE ADVANCEMENT
OF ANALYTICAL CHEMISTRY

Publication Committee

E. B. HUGHES, D.Sc., F.I.C.

F. W. F. ARNAUD, F.I.C.

A. L. BACHARACH, M.A., F.I.C.

R. C. CHIRNSIDE, F.I.C.

H. E. COX, D.Sc., Ph.D., F.I.C.

BERNARD DYER, D.Sc., F.I.C.

F. W. EDWARDS, F.I.C.

G. D. ELSDON, D.Sc., F.I.C.

B. S. EVANS, M.B.E., M.C., D.Sc., F.I.C.

E. HINKS, M.B.E., B.Sc., F.I.C.

G. ROCHE LYNCH, O.B.E., M.B.,
B.S., D.P.H., F.I.C.

G. W. MONIER-WILLIAMS, O.B.E.,
M.C., Ph.D., F.I.C.

A. MORE, I.S.O., A.R.C.S.,
A.R.T.C., F.I.C.

J. R. NICHOLLS, B.Sc., F.I.C.

W. H. ROBERTS, M.Sc., F.I.C.

W. H. SIMMONS, B.Sc., F.I.C.

ERIC VOELCKER, A.R.C.S., F.I.C.

K. A. WILLIAMS, B.Sc., F.I.C.

Hon. Secretary:

L. EYNON, B.Sc., F.I.C.

Hon. Treasurer:

G. TAYLOR, F.I.C.

Editor: C. AINSWORTH MITCHELL, M.A., D.Sc., F.I.C.

Secretary and Assistant Editor: J. H. LANE, B.Sc., F.I.C.

Abstractors:

S. G. CLARKE, Ph.D., B.Sc., A.I.C.

E. B. DAW, B.Sc., A.I.C.

J. GRANT, Ph.D., M.Sc., F.I.C.

D. G. HEWER, B.Sc.

A. O. JONES, M.A., F.I.C.

J. W. MATTHEWS, Ph.D., F.I.C.

E. M. POPE, B.Sc.

F. A. ROBINSON, M.Sc.Tech., F.I.C.

W. R. SCHOELLER, Ph.D., F.I.C.

D. R. WOOD, F.I.C.

VOL. 65

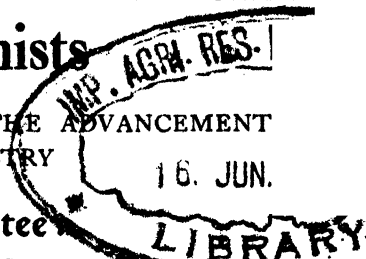
1940

PUBLISHED FOR THE SOCIETY BY

W. HEFFER & SONS LTD.

4, PETTY CURY, CAMBRIDGE, ENGLAND

1940



ERRATA:

VOL. 65, 1940:

- p. 27. Delete last line of "Official Appointments," i.e. Lincs. County, etc.
- p. 411. In list of Official Appointments for "Westmorland County Council, Kendal Borough, W. H. Roberts" read "W. H. Roberts as Public Analyst for that part of the County of Westmoreland within the boundaries of the Borough of Kendal."
- p. 519. Line 12 of Abstract on "Estimation of Phosphorus." For "about 0.4 mg. of phosphorus" read "not more than 0.4 mg. of phosphorus."
- p. 530. 2nd column: 7th line from bottom. For "J. Chem. Soc." read "J. Amer. Chem. Soc."
- p. 596. Last line but two of the Note: for " $\pm 0^\circ \text{C.}$ " read " $\pm 0.1^\circ \text{C.}$ "

INDEX TO VOLUME 65

INDEX TO NAMES

* Denotes authors of original papers and notes that have been published in *The Analyst*.

A

- Acree, F., Jr. See LaForge, F. B.
- Adickes, F. See Lang, O. St. A. K.
- *Agnew, W. J. The cyanometric determination of cobalt and nickel, 643.
- Ajtai, M. See Szebelledy, L.
- Aldred, P. A. See Koschura, W.
- Allen, N., Charbonnier, H. Y., and Coleman, R. M. Quantitative determination of certain polyalcohols in presence of each other, 575.
- Allen, R. J. L. Estimation of phosphorus, 519.
- Almquist, H. J., and Others. Identification of the rice factor, 573.
- Anderson, A. K. Essentials of Physiological Chemistry (Review), 76.
- Anderson, E. B. Review of Mason's *Fat, Total Solids and Moisture*, 75.
- *Andrew, R. L., and Neubauer, L. G. Succinic acid in beer, 455.
- Andrews, E. D. See Moir, G. M.
- Anslow, W. K., and Raistrick, H. Molecular constitutions of catenarin and erythroglaucon, 574.
- Anslow, W. K., Breen, J., and Raistrick, H. Studies on the biochemistry of micro-organisms. Emodic acid and ω -hydroxyemodin, metabolic products of *Penicillium cyclopium* Westling, 470.
- Appleyard, F. N., and Lyons, C. G. Practical Pharmaceutical Chemistry, 4th Ed. (Review), 436.
- Archbold, H. K. Sucrase activity in the barley plant, 520.
- Arnaud, F. W. F. Appointed Public Analyst for the Boroughs of Bromley, Bexley and Erith, 27; for the Borough of Dartford, 161; for the Borough of Gillingham (Kent) and the Urban District of Chislehurst and Sidcup, 411.
- Report of the County Analyst for Kent for Fourth Quarter, 1939, 221.
- Aschehoug, E., and Vesterhus, R. Significance of the bacterial count in determining the freshness of herring and brisling, the raw materials of kippered herrings and canned brisling, 417.

- Atherton, D., and Meara, M. L. Component fatty acids and glycerides of some *Myristica* fats, 516.
- Fatty acids and glycerides of solid seed fats. IX. *Mimusops Heckelii* (Baku) kernel fat, 517.
- Aufrecht, W. See Feigl, F.
- Auguste, G. Indirect volumetric determination of alkaloids, 110.
- Aull, J. C., and Kinard, F. W. Determination of tungsten in biological materials, 660.
- *Austin, G. J. A new group separation for the quantitative and qualitative analysis of phosphates, 335.
- Ayres, G. H., and Vienneau, E. M. Stability of peroxidised titanium solutions, 379.

B

- Bacharach, A. L. Review of Bernal's *The Social Function of Science*, 140.
- Review of Fearon's *An Introduction to Biochemistry*, 2nd Ed., 387.
- Review of Findlay's *Chemistry in the Service of Man*, 5th Ed., 535.
- Vitamin-free diets for animal experiments, 573.
- Bachman, C. Photometric determination of oestrogens. I. Modified Kober reaction for determining the total oestrogens in a mixture of oestrogenic steroids. II. New colour reaction for oestriol, 118.
- Bagchi, K. N., Ganguly, H. D., and Sirdar, J. N. Lead-content of human hair, 471.
- Bagnall, D. J. T. Appointed Public Analyst for the County Borough of Kingston-upon-Hull, 566.
- Bagnall, H. H. Report of the Birmingham City Analyst for 2nd quarter, 1940, 565; for the year 1939, 597.
- *Baker, A. Z., and Wright, M. D. Biological assay of vitamin D, 326.
- Baker, G. W. Report of the Government Analyst for Palestine for 1938, 166.
- Baker, W. O. See Caley, E. R.

- Balanescu, G., and Motzoc, M. D.** Determination of aluminium in presence of iron and phosphoric acid, 188.
- Baldwin, E., and Bell, D. J.** The glycogen of *Helix pomatia*, 468.
- Ballaban, H. E.** 1-2-Diaminoanthraquinone-3-sulphonic acid as a reagent for copper, 134.
- Ballentine, R.** See Taylor, A. R.
- Bandier, E.** Quantitative estimation of nicotinic acid in urine, 182.
- Banti, R. F.** See Marenzi, A. D.
- Barkan, G., and Walker, B. S.** Estimation of serum iron and pseudohaemoglobin iron with o-phenanthroline, 659.
- *Barraclough, A.** See Sutton, R. W.
- Barton, C. J.** See Yoe, J. H.
- Basu, U. P., and Mazumdar, A.** Keeping properties of *Hydnocarpus Wightiana* oil and its derivatives, 465.
- Baumgartner, J. G., and Knock, G. G.** Stability of bacteria in relation to pH, 371.
- Beacham, L. M.** Rapid method for the determination of chlorides in tomato products, 109.
- Beadle, D. G.** Examination of dusts from lungs produced by mining and other processes, 472.
- Beamish, F. E.** See Rogers, W. J.
- See also Seath, J.
- Beck, G.** Microchemistry of the rare earths, 382.
- Becker, E.** See Berg, R.
- Becker, E., and Kartos, R. F.** Vitamin C content of honey, 470.
- Bedford, C. L.** See Joslyn, M. A.
- Belcher, R., and Wheeler, R. V.** Studies in the composition of coal: Extraction of coal with quinoline, 614.
- Bell, D. J.** See Baldwin, E.
- Benedetti-Pichler, A. A., and Paulson, R. A.** Quantitative micro-determinations with the use of ordinary analytical balances, 69.
- Bennekou, J.** See Schou, S. A.
- Bennison, E. W., and Norris, F. W.** Pectic substances of plants. VI. Relation between jelly strength, viscosity and composition of various pectins, 67.
- Berg, R., and Becker, E.** Determination of cerium by means of 8-hydroxyquinoline, 312.
- Indo-oxine, a new precipitant for metals, 378.
- Berger, K. C., and Truog, E.** Determination of boron in soils and plants by means of quinalizarin, 123.
- Bergmann, E., Farkas, L., and Sandler, L.** Separation of *para*-hydrogen from oxygen and carbon monoxide, 244.
- Bergmann, M.** See Fruton, J. S.
- See also Ing, H. R.
- Bergmann, M., and Stein, W. H.** Naphthalene- β -sulphonic acid as a reagent for amino acids, 237.
- Bernal, J. D.** The Social Function of Science (Review), 140.
- Bernhardi, K., and Schoenheimer, R.** Inertia of highly unsaturated fatty acids in the animal, investigated with deuterium, 466.
- Bernhart, F. W.** Molecular weight of egg albumin, 236.
- Berrigan, D.** See Nelson, C. I.
- Bertrand, D.** Determination of small quantities of molybdenum in biological materials, 177.
- Besazza, V. L.** Adsorption of malachite green by clays and allied materials, 670.
- Biedebach, F., and Wiegand, H.** Detection of novocaine and identification of pantocaine, 173.
- Binkley, S. B., and Others.** Constitution of vitamin K₂, 524.
- Biroh, T. W., and György, P.** Physicochemical properties of the factor (vitamin H) curative of egg white injury, 117.
- Birnbaum, N., and Edmonds, S. M.** Determination of uranium and copper with the aid of the silver reductor, 313.
- Bisanz, R., and Kroupa, F.** Determination of fluorine and silica in the hot-springs of Gastein, 189.
- *Blackie, W. J., and Flemons, G. F.** Modification of the stirring device of the Hortvet cryoscope, 26.
- Block, R. J.** Basic amino acid content of human serum proteins. Influence of the ingestion of arginine on the composition of the serum proteins, 303.
- Block, R. J., and Bolling, D.** Micro-estimation of threonine, 72.
- Blumenthal, S.** See Fiene, F.
- Bodnár, J., and Török, T.** Photometric estimation of silicic acid in biological substance, 240.
- Bodnár, J., Szép, Ö., and Cieleaszy, V.** Rapid method for the accurate estimation of minute quantities of arsenic in biological material, 379.
- Boisson, R.** Determination of lactic and pyruvic acids by means of periodic acid, 473.
- Bolliger, A.** Volumetric determination of acridines with methylene blue, 423.
- Bolling, D.** See Block, R. J.
- Bond, G. W.** Application of hydrogen ion concentration to boiler water treatment, 429.
- Bonjean, P.** See Courtois, J.
- Booth, R. G.** Thiochrome method for the estimation of aneurin, with survey of the aneurin content of wheats, 610.
- Bose, P. K.** See Dutt, P.
- Bose, P. K., and Dutt, S. N.** Oil from the fruit of *Ferula alliacea*, 421.
- Bower, C. A., and Truog, E.** Manganese method for determination of base exchange capacity of soils and other materials, 614.
- Boyland, E.** See Chibnall, A. C.
- Bozain, M.** See McIntyre, G. H.
- Bracaloni, L.** Presence of *l*-tyrosine in the alcoholic extract of egg yolk, 298.
- Bradbury, F. R., and Edwards, E. G.** Volumetric determination of ferric iron by means of mercurous nitrate, 433.
- Bradley, H. B.** See Derrett-Smith, D. A.
- Bradstreet, R. B.** See Lewis, J. B.
- Brady, O. L.** Review of Weeks's *Discovery of the Elements*, 4th Ed., 321.
- Brame, J. S. S.** Obituary of F. G. Edmed, 391.
- Brand, E., and Kassell, B.** Determination of reducing groups with porphyrindin, with special reference to egg albumin, 425.
- Bray, R. H.** See Dickman, S. R.
- Breen, J.** See Anslow, W. K.
- Brindle, H.** Phenol and eucalyptus ointments, 608.
- The iodine value of soft paraffins, 409.

- *Britton, H. T. S. Universal buffer mixture, 220.
- Britton, H. T. S., and Königstein, M. Potentiometric determination of hydroxylamine and hydrazine in alkaline solution, 532.
- *Britton, H. T. S., and Phillips, L. The potentiometric estimation of glucose with potassium ferricyanide in sodium carbonate solution, 149.
- *— The potentiometric titration of glucose with alkaline tartrate solutions of copper, including Fehling's solution, 18.
- Brodie, W. R. Chemical Spectroscopy (Review), 137.
- Brooks, J., and Pace, J. Manometric estimation of nitrite in solution and in tissues, 468.
- Brooks, J., Haines, R. B., Moran, T., and Pace, J. The function of nitrate, nitrite and bacteria in the curing of bacon and ham, 355.
- Brown, J. B. See Shinowara, G. Y.
- Brown, R. A. See Emmett, A. D.
- Browning, B. L. See Stillings, R. A.
- Brückmann, G., and Zondek, S. G. Improved method for the estimation of non-haemin iron, 659.
- Brüning, A. Toxicological detection of cocaine, 427.
- Brunner, K., and Neugebauer, H. Determination of the alkaloids in preparations of *Berberis vulgaris* and *Berberis agrifolium*, 47.
- Buckler, E. J. See Newton, A.
- Budhiraja, K. L. Report of the Chief Chemist for the State of Patiala for 1939-40, 652.
- Bueding, E., and Wortis, H. Stabilisation and determination of pyruvic acid in blood, 426.
- Buell, M. V. Method of ashing soft tissues preliminary to the determination of cations, 53.
- Bukatsch, F. Estimation of vitamin C by means of the Zeiss step-photometer, 241.
- Burchard, C. See Müller, E.
- Burr, G. O. See Kass, J. P.
- Burt, G. M. Report of the Building Research Board for 1939, 598.
- Burton, D., and Robertshaw, G. F. Sulphated Oils and Allied Products—Their Chemistry and Analysis (Review), 318.
- Unsaponifiable matter in sulphated oils and fatty alcohols. Committee on oils, fats and waxes. No. XIII, Part I, 615.
- Butler, A. M. See Talbot, N. B.
- Buxton, L. O. See Simons, E. J.
- Buxton, L. O., and Kapp, R. Purification of high molecular weight fatty esters, 528.
- C**
- Caley, E. R., and Baker, W. O. Composition of lithium and potassium salts precipitated by uranyl acetate reagents for sodium, 131.
- Cardoso, H. T. See Cole, H. I.
- Carpenter, P. See Levine, M.
- Carson, W. L. Luximeter: Construction and application, 534.
- Cartwright, L. C., and Epstein, S. S. Routine determination of the bacterial content of paper-board, 612.
- Castiglioni, A. Determination of piperazine, 422.
- Chakravarti, S. M. Report of the Chemical Examiner for the United and Central Provinces of India for 1939, 601.
- Challis, H. J. G. See Friend, J. N.
- Chalmers, F. G. D. Appointed Agricultural Analyst for the County Council of Warwickshire, 411.
- Appointed Deputy Public Analyst for the Borough of Rugby, 27; Public Analyst for the Borough of Nuneaton, 161; Additional Public Analyst for the Borough of Hereford, 411.
- Chandlee, G. O. See Hayes, J. R.
- Charaux, C., and Rabaté, J. Study of *Centaurea scabiosa*, 302.
- Charbonnier, H. Y. See Allen, N.
- Cheffell, H. See Panouse-Digeaud, L.
- Cheramy, P. Detection of chloralose in urine, 471.
- Cheramy, P., and Lemos, A. Toxicology of sodium and potassium permanganates, 120.
- Cheramy, P., and Papavassilou, M. Extraction of alkaloids with acetone, 243.
- Chernoff, L. H. Determination of indole by a modification of Ehrlich's reaction, 465.
- Chibnall, A. C., and Others. Glutamic acid of normal and malignant tissue proteins, 520.
- Chirnside, R. C. Review of Sand's *Electrochemistry and Electrochemical Analysis*, Vol. II, 620.
- Review of Thompson's *Theoretical and Applied Electrochemistry*, 3rd Ed., 193.
- *Chirnside, R. C., Dauncey, L. A., and Proffitt, P. M. C. The determination of aluminium, magnesium or beryllium in nickel alloys, 446.
- Cholnoky, L. Quantitative estimation of coloured components in paprika, 232.
- Christiansen, B. E., and Faer, J. F. Wet combustion method for determination of carbon and of oxygen consumed, 64.
- Christiansen, W. G. See Jurist, A. E.
- Cioekdagl, F. See Heiduschka, A.
- Ciolesky, V. See Bodnár, J.
- Cimerman, C., and Wenger, P. Micro-determination of zinc by the hydroxyquinoline method. IV. Separation from manganese, iron, bismuth, mercury, arsenic and antimony, 381.
- Clark, W. Photography by Infra-red: Its Principles and Applications (Review), 480.
- Clayton, W. Review of Weiser's *Colloid Chemistry*, 536.
- Clegg, G. G. Examination of damaged cotton by the Congo red test: Further developments and applications, 475.
- Coblentz, J. M. See Levine, M.
- Cochran, J. A. Lavoisier (Review), 320.
- Cohen, B. R. See Wikoff, H. L.
- Cohen, I. Evaluation of the activity of powders of *Veratrum viride* by the *Daphnia* method, 184.
- Cole, H. I., and Cardoso, H. T. Analysis of chaulmoogra oils. *Hydnocarpus anthelmintica* oil and *Taraktogenos Kurrii* (chaulmoogra) oil, 231; III. *Hydnocarpus wightiana* oil, 45.
- Coleman, R. M. See Allen, N.
- Collin-Russ, A. Human foot perspiration and upper boot leather, 659.

- Collins, J. V.** Report of the Government Analyst for Ceylon for 1939, 652.
- Colman, H. B.** See Simons, E. J.
- *Colson, A. F.** The micro-titrimetric determination of carbon dioxide in carbonates, 638.
- *Common, R. H.** The phytic acid content of some poultry feeding stuffs, 79.
- Comrie, A. A. D.** Colorimetric determination of the preservative value of hops. Standard colour values of some hybrid hops, 516.
- Connah, F. E.** Report of the Government Analyst for Queensland for 1938-39, 164.
- Constantinesco, O.** See Ionesco-Matiu, A.
- Conway, E. J.** Micro-diffusion Analysis and Volumetric Error (Review), 254.
- Coops, J.** See Mendel, H.
- Cordebard, M. H.** Volumetric determination of organic substances by chromic oxidation: Nitro-chromic solutions, 123.
- Courtois, J., and Bonjean, P.** Volumetric determination of sulphate in water, 121.
- Coward, K. H., and Kassner, E. W.** Estimation of vitamin D in food substances containing phosphorus, 523.
- Cox, H. E.** Hair dyes. II. The functions and reactions of phenols, 393.
- Review of Thorpe and Whiteley's *Thorpe's Dictionary of Applied Chemistry*, 4th Ed., 583.
- Crandall, L. A.** Method for the estimation of acetone bodies in blood, applicable also to the estimation of small amounts of mercury, 467.
- Cumming, A. C., and Kay, S. A.** A Text-book of Quantitative Chemical Analysis. 7th Ed. (Review), 138.
- Cutts, H. E.** Obituary of T. S. Gladding, 391.
- D**
- Daglish, C., and Wokes, F.** Chemical assay of powdered ergot, 50.
- Dahl, O.** Characterisation of starch from various sources by means of electrophoretic fractionation, 362.
- Dalby, G.** See Hoffman, C.
- Danjo, M.** See Takano, M.
- Darby, W. J., and Day, P. L.** Blood sugar levels in rats receiving the cataractogenic sugars galactose and xylose, 520.
- Darnell, M. C., Jr., and Walker, V. S.** Determination of sodium in biological fluids, 478.
- Dasa Rao, C. J.** See Vidyarthi, N. L.
- *Dauncey, L. A.** See Chirside, R. C.
- Davenport, J. E.** See Evans, R. N.
- Davidson, D.** Qualitative test for organic compounds containing oxygen, 306.
- Davies, A. W., and Moore, T.** Vitamin A content of cheese, 58.
- Davis, T. L., and Green, W. P.** New reaction of formic acid, 528.
- Davis, V. E.** See Lewis, D. T.
- Day, P. L.** See Darby, W. J.
- Deahl, N.** See Lynch, H. J.
- Deb, N. C.** See Dutt, P.
- *De Giacomini, R.** The determination of tin in foodstuffs by means of dithiol, 216.
- de la Rivière, E. D.** Practical methods for sterilisation of water consumed by evacuated populations, 429.
- Delga, J.** New colour reaction of phenarsazine chloride, 309.
- Reactions of Nessler's reagent with aqueous solutions of mustard gas and lewisite, 246.
- Delory, G. E.** See King, E. J.
- Denoël, A.** Determination of bromides in presence of chlorides, 314.
- Derrett-Smith, D. A., and Bradley, H. B.** Machine for testing the fastness to rubbing of dyed materials, 534.
- Deturk, E. E.** See Stubblefield, F. M.
- Dhawan, C. L.** See Hoon, R. C.
- Dickhart, W. H.** Behaviour of olive oil and other oils with antimony trichloride, 421.
- Dickinson, D.** Determination of chromates in sewage liquors, 409.
- *Dickinson, D., and Weir, E. M.** A rapid stability test for sewage effluents, 289.
- Dickman, S. R., and Bray, R. H.** Removal of phosphoric acid from hydrogen peroxide, 477.
- Diemair, W., and Koch, J.** Estimation of sulphur compounds in vegetables, 418.
- Discherl, W.** Colour reaction for the detection of the equal of mare's urine, 368.
- Dixon, F.** Appointed Deputy Public Analyst for the Urban District of Brierley Hill, 411.
- Dixon, S.** Report of the Public Analyst for Cardiff City for 1938, 28.
- Doisy, E. A.** See Binkley, S. B.
- Donath, W. F.** See Sjollem, B.
- Donau, J.** Micro-electrolytic determinations in small volumes, 134.
- Donen, I.** Rôle of sorbitol in the carbon-metabolism of the Kelsey plum. I. Changes in chemical composition during growth and storage, 178.
- Dorsey, N. E.** Properties of Ordinary Water Substance (Review), 620.
- Dott, D. B.** Tests for tannic acid, 233, 424.
- Duke, F. R., and Smith, G. F.** Rapid qualitative test for alcoholic hydroxyl groups, 527.
- *Dunbar-Poole, A. G.** Antimony in tin-antimony oxides, 453.
- Dunn, R., Inouye, K., and Kirk, P. L.** Microscopy of the amino acids and their compounds, 252.
- Dutt, P., Deb, N. C., and Bose, P. K.** Mesuol: the bitter principle of *Mesua ferrea*, 466.
- Dutt, S. N.** See Bose, P. K.
- Dyer, B.** Appointed Joint Public Analyst for the Borough of Ilford, 27; for the Borough of Wanstead and Woodford, 161.
- Obituary of John White, 325.
- E**
- Eakin, R. E.** See Snell, E. E.
- *Edisbury, J. R.** Spectrophotometric assay of vitamin A, with special reference to margarine, 484.
- Edmonds, S. M.** See Birnbaum, N.
- Edwards, E. G.** See Bradbury, F. R.
- Edwards, F. W.** Review of Radley's *Starch and Its Derivatives*, 481.
- Edwards, S. J.** Diagnosis of *Streptococcus mastitis* by cultural methods, 62.
- Egriwe, E.** Tests for cadmium and magnesium, 248.

- Egell, B. D., and Gerhardt, F.** Use of ascorbic acid as a substrate in oxidase measurements, 468.
- Egloff, G.** Physical Constants of Hydrocarbons, Vol. II (Review), 585.
- Eichler, H.** Identification of groups of dyestuffs by their reaction with ferrous hydroxide, 431.
- Ellas, A.** Hydrogen sulphide in corned beef, 605.
- Elliott, E. C.** Determination of sodium in presence of other metals, 616.
- Ellis, G.** See Voris, L.
- Ellis, R. H.** Appointed Public Analyst for the Borough of Swindon, 161.
- El Sadr, M. M., Macrae, T. F., and Work, C. E.** Estimation of riboflavin. I. New biological method, 522.
- Elson, G. D.** Review of Hilditch's *The Chemical Constitution of Natural Fats*, 319.
- *Elsworth, M.** See Garner, W.
- Elvehjem, C. A.** See Hove, E.
- Emmerie, A.** Colorimetric determination of tocopherol (vitamin E). IV. Quantitative determination of tocopherol in oils after saponification, 370.
- Emmett, A. D., Kamm, O., and Sharp, E. A.** Vitamin K activity of 4-amino-2-methyl-1-naphthol and 4-amino-3-methyl-1-naphthol, 305.
- Emmett, A. D., Peacock, G., and Brown, R. A.** Estimation of aneurin by a modification of the Melnick-Field method, 661.
- Emte, W.** See John, W.
- Engelder, C.** Calculations of Quantitative Analysis (Review), 674.
- Englis, D. T.** See Shannon, W. J.
- Ephraim, F.** Inorganic Chemistry, 3rd English Ed. (Review), 584.
- Epstein, S. S.** See Cartwright, L. C.
- Epstein, S. S., and Snell, F. D.** Thermal death time of micro-organisms encountered in the brewing industry, 524.
- *Etheridge, A. T.** The determination of phosphorus in titanium steels, 496.
- Evans, B. S.** Review of Ephraim's *Inorganic Chemistry*, 3rd English Ed., 584.
- Review of Mellor's *Modern Inorganic Chemistry*, 9th Ed., 479.
- Evans, E. A.** See Krebs, H. A.
- Evans, J.** Appointed Public Analyst for the Borough of Scunthorpe, 161.
- Evans, R. C.** An Introduction to Crystal Chemistry (Review), 75.
- Evans, R. N., Davenport, J. E., and Revukas, A. J.** Micro-gravimetric determination of active hydrogen by the Grignard reagent, 533.
- *Evans, T.** The net calorific value of coals, 352.
- F**
- Facer, J. F.** See Christiansen, B. E.
- Fairbrother, R. W.** A Textbook of Bacteriology, 3rd Ed. (Review), 673.
- Farkas, L.** See Bergmann, E.
- Fearon, W. R.** An Introduction to Biochemistry, 2nd Ed. (Review), 387.
- Feigl, F., and Aufrecht, W.** Detection of barium sulphate by means of spot reactions, 251.
- Ferguson, A.** Review of Evans's *An Introduction to Crystal Chemistry*, 75.
- Fernholz, E., and Moore, M. L.** Isolation of α -spinasterol from alfalfa, 60.
- Fernholz, E., and Staveland, H. E.** Brassicasterol, the characteristic sterol of rapeseed oil, 572.
- Ferrey, G. J. W.** Determination of mercury in mercurochrome, 607.
- Field, H.** See Melnick, D.
- Fiene, F., and Blumenthal, S.** Handbook of Food Manufacture (Review), 255.
- *Fife, J. G.** Studies in internal electrolysis. V. The determination of small quantities of copper in presence of cadmium, 562.
- Findlay, A.** Chemistry in the Service of Man, 5th Ed. (Review), 535.
- Fischer, H., and Leopoldi, G.** Colorimetric determination of small amounts of lead by means of dithizone, 432.
- Fischer, R.** Detection of ammonia, calcium and strontium with organic nitro compounds, 135.
- Flagg, J. F., and Furman, N. H.** Qualitative reactions of salicylaldoxime derivatives with inorganic ions, 668.
- *Flemons, G. F.** See Blackie, W. J.
- Flint, J. W.** Appointed Deputy Public Analyst for the Boroughs of Beckenham, and Bromley, 161.
- Flügel, J.** See Komm, E.
- Foerster, P.** Determination of lead in tin coatings, 476.
- Ford, W. P., and Others.** Micro-determination of bromine in foodstuffs, 617.
- Foster, L. S.** Reaction of gallium with perchloric acid, 68.
- Fox, J. J.** Review of Brode's *Chemical Spectroscopy*, 137.
- Review of Hibbin and Teller's *The Raman Effect and its Chemical Applications*, 192.
- Frankenthal, L.** Methylene blue reducing system of Palestine orange peels investigated by Thunberg system, 662.
- Fraps, G. S., and Fudge, J. F.** Determination of iodine in soils, plant material and waters, 305.
- Fraps, G. S., Kemmerer, A. R., and Greenberg, S. M.** Adsorption method for the determination of pure carotene, 609.
- Determination of carotene in presence of lycopene, 608.
- *Friend, J. N., and Hall, D. A.** Estimation of praseodymium and neodymium in solution from their adsorption spectra, 144.
- Friend, J. N., Vallance, R. H., and Challis, H. J. G.** Phosphate separation in qualitative analysis, 578.
- Fruton, J. S., and Bergmann, M.** Activation of papain, 303.
- Fuchs, W., Ishler, N. H., and Sandhoff, A. G.** Determination of hydroxyl groups with the Grignard reagent, 663.
- Fudge, J. F.** See Fraps, G. S.
- Fuller, P. N.** Review of Egloff's *Physical Constants of Hydrocarbons*, Vol. II, 585.
- Furman, N. H.** See Flagg, J. F.
- G**
- Gade, G. N.** See Townsend, F. E.
- Ganguly, H. D.** See Bagchi, K. N.
- *Garner, W.** A laboratory textile scouring apparatus, 563.
- An alkaline ageing test for textile oils, 219.

- *Garner, W., and Elsworth, M. The peroxide accumulation rate of oils—relationship to Mackey test results and oxidation on textiles, 347.
- Gaspard, R., and Serrure, G. Determination of water in ether, 73.
- Gatti, C., and Knallinsky, A. Tillmann reaction for vitamin C in plant extracts, 369.
- *Gee, G. N. The detection of urea in urine stains on carpets, 25.
- Geiger, E., and Müller, G. Microchemical determination of the reducing power of cellulose, oxycellulose and hydrocellulose (micro-copper, micro-ferricyanide and micro-iodine numbers), 618.
- Georgi, E. A. Analysis of rosin size, 614.
- Gerhardt, F. See Egell, B. D.
- Gershenfeld, L., and Perlstein, D. Preservatives for preparations containing gelatin, 62.
- Getz, C. A. See Smith, G. F.
- Ghosh, B., and Guha, B. C. Concentration of ascorbigen from cabbage, 523.
- Ghosh, S. K. See Quadrati-Khuda, M.
- Gilbert, G. R., and Tannich, R. E. Detection of aromatics in air, 666.
- Ginsburg, J. M. Chemical effect on lead arsenate of certain salts which may be present in soil and spray waters, 613.
- Glasscock, H. H., and Wain, R. L. Distribution of manganese in the pea seed in relation to marsh spot, 372.
- Glimm, E., Wittmeyer, H., and Jahn-Held, W. Thermo-stability of fats, 301.
- Goepfert, G. J. Micro-determination of succinic acid, 580.
- Goldberg, W., and Walsh, E. O'F. Ascorbic acid content of rose hips, 304.
- Golumbic, C., and Mattill, H. A. Oxidation of vitamin E, 573.
- Goodhue, L. D., and Haller, H. L. Determination of deguelin in derris and cubé, 184.
- Goodson, J. A. Pelletierine of commerce, 422.
- Gopala Rao, G., and Sundara Rao, W. V. Mechanism of the microbial oxidation of ammonia, 524.
- Gordon, N., and Trout, W. E., Jr. Introductory College Chemistry, 2nd Ed. (Review), 537.
- Gornall, A. G., and Hunter, A. Behaviour of some uramido-acids in the nitrous acid method for the determination of amino-nitrogen, 467.
- Goss, M. J. See Phillips, M.
- Gourevitch, A. Separation of the components of a mixture by fractional centrifuging, 136.
- Grabowska, G. See Iwanowski, W.
- Graft, J. H., Schlosser, M. A., and Nihlen, E. K. Cross sections of pulp fibres, 670.
- Grangaud, R. New method for washing and isolation of barium sulphate in the micro-determination of sulphur, 316.
- Grant, J. See Radley, J. A.
- Grant, J. B. Report of the Director of the All-India Institute of Hygiene and Public Health for 1938, 225.
- Gray, A. G. See Holt, M. L.
- Green, W. P. See Davis, T. L.
- Greenberg, S. M. See Frape, G. S.
- Greenstein, E. J. Fertiliser problems in vegetable production, 416.
- Greeves, W. F. Appointed Deputy Agricultural Analyst to the County Council of East Suffolk, 411.
- Appointed Deputy Public Analyst for the Counties of Norfolk and East and West Suffolk, and the Boroughs of Great Yarmouth and Lowestoft, 566.
- *Gridgeman, N. T., Lees, H., and Wilkinson, H. Estimation of vitamin D in margarine, 493.
- Griebel, C., and Hess, G. Iodine absorption of honey and artificial honey, 299.
- Griffin, D. B. See Hall, R. T.
- Griffiths, J. G. A. Review of Dorsey's *Properties of Ordinary Water Substance*, 620.
- Grimmett, R. E. E. Report of Chemistry Section, New Zealand Dept. of Agriculture for 1938-39, 33.
- Grimmett, R. E. E., and I. G. McIntosh. Arsenic in soils and waters in the Waitapu Valley, New Zealand, 613.
- Grossfeld, J. Determination of hydrocarbons in unsaponifiable matter, 375.
- Grossfeld, J., and Payler, E. Determination of formic acid in foodstuffs, 168.
- Grossman, M. J. See Wikoff, H. L.
- Grumer, J. See Molnar, N. M.
- Guénin, J. See Péronnet, M.
- Guha, B. C. See Ghosh, B.
- See also Pal, J. C.
- See also Sen-Gupta, P. N.
- Guillaume, A., and Nicolas, Y. Stability of calcium hypochlorites, 130.
- Gunde, B. G., and Hilditch, T. P. Mixed unsaturated glycerides of liquid seed fats. Some "non-drying" oils, 364.
- Guthrie, F. C. Modified iodate method for the determination of barium, 434.
- Guthrie, F. C., and Nance, J. T. Revision of Cumming and Kay's *A Text-book of Quantitative Chemical Analysis*, 7th Ed., 138.
- György, P. Curative factor (vitamin H) for egg white injury with particular reference to its presence in different foodstuffs and in yeast, 117.
- See also Birch, T. W.
- György, P., Kuhn, R., and Lederer, E. Attempts to isolate the factor (vitamin H) curative of egg white injury, 117.

H

- Haines, R. B. See Brooks, J.
- *Hall, D. A. See Friend, J. N.
- Hall, P. E. Analyses of certain South African woods, with special reference to their use as producer-gas generator fuels, 577.
- Hall, R. T., Holcomb, J. H., Jr., and Griffin, D. B. Separation and determination of isomeric menthols, 518.
- Hall, W. T. Revision of Moore's *A History of Chemistry*, 3rd Ed., 386.
- Haller, H. L. See Goodhue, L. D.
- Hallinan, F. J. Colorimetric determination of free chlorine in effluents, 578.
- Halverson, J. O., and Sherwood, F. W. Vitamin A activity and vitamin B₁ content of soyabans and cowpeas, 470.

- ***Hamence, J. H.** A new test for the detection of molybdenum and tungsten, 152.
- Appointed Joint Public Analyst for the Borough of Ilford, 411.
- Hammond, J. W., and MacIntire, W. H.** Thorium nitrate micro-titration of fluorine in aqueous and alcoholic systems, 618.
- Haripe, J. V.** Colorimetric semi-micro-determination of arsenic in urine, 54.
- Harper, S. H.** Active principles of leguminous fish-poison plants. IV. Isolation of malaccol from *Derris malaccensis*, 472.
- Harral, J. C.** The determination of cocoa alkaloids in confectionary, 408.
- Harris, L. J.** Vitamins (Review), 139.
- Harris, L. J., and Raymond, W. D.** Estimation of nicotinic acid in urine, 183.
- Harrison, C. J., and Roberts, E. A. H.** Fermentation process in tea manufacture. IV. Tea tannin and its fermentation products, 46.
- Harrison, D. C., and Mellanby, E.** Phytic acid and the rickets-producing action of cereals, 113.
- Hart, D., and Meyrowitz, R.** Detection of thiocyanate, iodide, bromide and chloride, 532.
- Hart, E. B.** See Hove, E.
- Hartmann, M., and Schlitter, E.** African arrow-poisons. I. *Adenium somalense* Balf. fil., 428.
- Haslam, J., and Roberts, R. F.** The iodimetric determination of alkali: An investigation of G. S. Smith's method, 399.
- Haskó, A.** Action of mustard gas on foods, 427.
- Hawkins, E. G. E., Jones, J. K. N., and Young, G. T.** Constitution of banana starch, 464.
- ***Hawley, H.** Cleaning Jena glass filters, 27.
- Neutralisation value of ghee butter fat, 606.
- Report of the Government Analyst for Madras for 1938-39, 601.
- * — The use of anhydrite as a desiccating agent, 644.
- Hayes, J. R., and Chandless, G. C.** Determination of bismuth by precipitation as quinaldine iodobismuthite, 129.
- Heiduschka, A., and Cicekdagi, F.** Milk fat from the Turkish buffalo, 297.
- Heiman, S.** See McNabb, W. M.
- Hendrey, W. B.** Estimation of soya-bean flour in sausage by determining non-fermentable sugars, 109.
- ***Hennes, J. R.** See Prideaux, E. B. R.
- Henry, K. M., Houston, J., and Kon, S. K.** Estimation of riboflavin. II. In milk: comparison of fluorimetric and biological tests, 522.
- Henville, D.** Report of Public Analyst for Metropolitan Borough of Stepney for 1938, 29.
- Herzfeld, E.** See Verda, A.
- Herzog, R.** See Sherman, C. S.
- Hess, G.** See Griebel, C.
- Hettche, O.** Silicic acid nutrient media, 243.
- Hewitt, J. T.** Obituary of Sir G. T. Morgan, 324.
- Hibben, J. H., and Teller, E.** The Raman Effect and its Chemical Applications (Review), 192.
- Hidaka, T.** Rice bran as a raw material of oil, 44.
- Hilditch, T. P.** Foreword to Burton and Robertshaw's *Sulphated Oils and Allied Products*, 318.
- The Chemical Constitution of Natural Fats (Review), 319.
- See also Gunde, B. G.
- Hilditch, T. P., and Maddison, L.** Composition of commercial palm oils. V. Partial separation of palm oils by crystallisation as an aid to the determination of the component glycerides, 517.
- Mixed unsaturated glycerides of liquid seed fats. II. Low temperature crystallisation of cottonseed oil, 556.
- Hilditch, T. P., and Murti, K. S.** Component acids and glycerides of some Indian ox depot fats, 555.
- Fat of land crabs (Seychelles Islands), 518.
- * — The use of thiocyanogen values in the determination of linoleic and linolenic acids and their glycerides, 437.
- Hilditch, T. P., and Pedelty, W. H.** Body fats of the pig. V. Component glycerides of perinephritic and outer back fats from the same animal, 569.
- Hilditch, T. P., and Saul, S.** Component glycerides of a typical cow milk fat, 606.
- Hills, G. M.** Thiochrome test for aneurin in urine as an index of nutritional level, 179.
- Hilly, W. W., and Wilson, D. T.** Determination of iodine in thyroid and its preparations by cerate oximetry, 174.
- Hind, H. G.** Colouring matters of *Penicillium carmino-violaceum* Biourge, with a note on the production of ergosterol by this mould, 470.
- Constitution of carviolin: a colouring matter of *Penicillium carmino-violaceum* Biourge, 470.
- Hind, H. Lloyd.** Brewing Science and Practice. Vol. II. Brewing Processes (Review), 482.
- Hinks, E.** Appointed Public Analyst for the Urban District of Esher and the Boroughs of Surbiton, and Malden and Coombe, 27; for the Urban Districts of Coulsdon and Purley, and Merton and Morden, and for the Borough of Wimbledon, 161; for the Boroughs of Kingston-upon-Thames and Mitcham, and the Urban District of Carshalton, 411.
- ***Hinton, C. L., and Macara, T.** The composition of some jam fruits and the determination of the fruit content of jams, 540.
- Hirst, F.** Report of the Director of the Fruit and Vegetable Preservation Research Station, Campden, for 1939, 511.
- Hiskey, C. F., and Meloche, V. W.** Determination of rhenium in molybdenum, 669.
- Hiskey, C. F., Springer, V. F., and Meloche, V. W.** Reduction of molybdate in the silver reductor, 69.
- ***Hitchen, O.** See Sutton, R. W.
- Hoar, T. P.** Hydrogen "swells" in canned fruits, 43.
- Hodgson, T. R.** Appointed Public Analyst for the Borough of Crewe, 161; for the Borough of Swinton and Pendlebury, 566.
- Hodson, A. Z., and Norris, L. C.** Fluorometric method for determining the riboflavin content of food-stuffs, 114.
- Hoffman, C., Schweitzer, T. R., and Dalby, G.** Determination of iron in bread and bread ingredients, 569.
- Hoffman, C. H.** See Tchitchibabine, A. E.
- Hogden, C. G.** See Robinson, H. W.
- Holcomb, J. H., Jr.** See Hall, R. T.

- Hollman, E. C. M. J.** Determination of the ascorbic acid content of tablets, 365.
- Holm, J. M., and Marloth, B. W.** The Bambarra groundnut or Njugo bean, 608.
- Holmstrom, J. E.** Records and Research in Engineering and Industrial Science (Review) 322.
- Holt, M. L., and Gray, A. G.** Volumetric determination of tungsten, 312.
- Holtz, P.** Combined ascorbic acid of animal tissue, 369.
- Hoon, R. C., and Dhawan, C. L.** Electrodialysis of soils. Influence of exchangeable bases on the recovery of manganese by electrodialysis, 526.
- Hopkinson, F. J.** See Wernimont, G.
- Hooshall, E. M.** Determination of sulphides in depilatories, 579.
- Determination of thioglycollates in depilatories, 667.
- Houston, J.** See Henry, K. M.
- Hove, E., Elvehjem, C. A., and Hart, E. B.** Effect of zinc on alkaline phosphatases, 572.
- Howe, D. E., and Mellon, M. G.** Colorimetric determination of iron with salicylaldoxime, 578.
- Hughes, L. E. C.** See Tweeney, C. F.
- Hunter, A.** See Gornall, A. G.
- Hunter, D.** Toxicology: Industrial Poisoning (Review), 139.
- Hynes, W. A.** See Yanowski, L. V.
- Hynes, W. A., and Yanowski, L. K.** Salts of complex cations applied to the microscopical detection of anions, 70.
- Jaffé, F. W. M.** Appointed Deputy Public Analyst for the County Borough of Wakefield, 566.
- Appointed Public Analyst for the Borough of Keighley, 27; for the Borough of Harrogate, 161; for the Borough of Batley and the Urban District of Castleford, 411.
- Jahn-Held, W.** See Glimm, E.
- *James, G. V.** The determination of members of the sulphanilamide group of drugs, 206.
- Janke, A., and Mikschik, E.** Micro-potentiometric method of formol titration, 434.
- Jensen, C. C.** See Schoch, T. J.
- Jensen, H. B., and With, T. K.** Vitamin A and carotenoids in the liver of mammals, birds, reptiles and man, 179.
- Jensen, K. E.** Estimation of foreign organisms in yeast, 62.
- John, W., and Emte, W.** The red oxidation products of the tocopherols, 242.
- Johnson, K. R.** See Mehlig, J. P.
- *Johnson, T. E.** See Kersey, R. W.
- Jones, A. O.** Review of *Annual Reports of the Progress of Applied Chemistry*. Vol. XXIV for 1939, 436.
- Jones, E. V.** Appointed Public Analyst for the Urban District of Brierley Hill, 411.
- Jones, J. K. N.** See Hawkins, E. G. E.
- Joslyn, M. A., and Bedford, C. L.** Enzyme activity in frozen vegetables: asparagus, 468.
- Jurist, A. E., and Christiansen, W. G.** Oxalate formation in ascorbic acid solutions, 184.

K

- I**
- Ichiyanagi, B.** See Kawakita, K.
- Ikert, B.** Determination of fluorine in wood (treated with fluoride preservative), 190.
- *Illing, E. T.** The detection and estimation of benzedrine, 3.
- Ing, H. R., and Bergmann, M.** Semi-micro-estimation of amino acids, 57.
- Inouye, K.** See Dunn, R.
- Insinger, J. A.** See Rowaan, P. A.
- Ionescu-Matiu, A., Popesco, C., and Constantinesco, O.** Micro-determination of ethyl alcohol in pharmaceutical products. Determination of the chromic index, 135.
- Irwin, J. O.** Estimation of riboflavin. III. Statistical analysis of the data, 522.
- Irwin, W. B.** See Smith, L. I.
- Ishler, N. H.** See Fuchs, W.
- Iwanowski, W., and Grabouska, G.** Determination of sugars in flours, 168.
- J**
- Jackerott, K. A., and Reimers, F.** Determination of nicotinic acid diethylamide and phthalic acid bis-diethylamide, 234.
- Jacob, A., and Todd, A. R.** *Cannabis indica*. Isolation of cannabidiol from Egyptian hashish. Observations on the structure of cannabinol, 466.
- Jaeger, W.** See Karrer, P.
- Kahler, H. L.** Interference of sulphite with the determination of sulphate by the tetrahydroxyquinone method, 434.
- Kaiser, E. P., and Parrish, W.** Immersion liquids for determination of refractive index of non-opaque minerals, 137.
- Kamm, O.** See Emmett, A. D.
- Kapp, E. M.** Micro-estimation of uronic acids, 580.
- Kapp, R.** See Buxton, L. O.
- Kardos, R. F.** Reducing power of unripe honey and honey from sugar-fed bees, 363.
- See also Becker, E.
- Karrer, P.** Blue alkali salts of α -phyloquinone (vitamin K₁) and similar compounds, 60.
- Karrer, P., Jaeger, W., and Keller, H.** Estimation of tocopherol in animal organs, 427.
- Karush, F.** See Talbot, N. B.
- Kass, J. P., and Others.** Thiocyanogen value of linolenic acid, 528.
- Kassall, B.** See Brand, E.
- Kassner, E. W.** See Coward, K. H.
- Kawakita, K., and Ichiyanagi, B.** Elimination of traces of carbon monoxide from hydrogen, 575.
- Kay, S. A.** See Cumming, A. C.
- Kaye, A.** Micro-determination of potassium. I. 533.
- Keiser, B.** Identification of 2-aminoethanol, 474.
- Keller, H.** See Karrer, P.
- Keller, H., and Möller, H.** Detection by Walkiewicz's method of the decomposition of meat, 418.

- Kemmerer, A. R.** See Fraps, G. S.
- Kent, M. L.** Quantitative analysis of plant tissues for lithium by the Ramage flame spectrographic method, 581.
- Kent-Jones, D. W.** See Ford, W. P.
- Keresztessy, J. C.** See Scudi, J. V.
- *Kersey, R. W., Maddocks, J. R., and Johnson, T. E.** The determination of small amounts of formaldehyde in air, 203.
- Kewley, J.** Review of Sachanen's *Conversion of Petroleum*, 671.
- Kieft, J. P.** Inactivation of the enzymes of gum arabic, 112.
- Kiffer, A. D.** See Lamprey, H.
- Kimura, W., and Tsurugi, J.** Arylation of oils and fats. Synthesis of tolyl stearic acid and its esters, 186.
- Kinard, F. W.** See Aull, J. C.
- King, E. J., and Delory, G. E.** Carbonate veronal buffer solution covering pH 7.5 to 10.7, 469.
- King, H.** Alkaloids of some *Chondrodendron* species and the origin of *Radix Pareirae Bravae*, 518.
- Kingsbury, A. N.** Report of the Director of the Federated Malay States Institute of Medical Research for 1938, 459.
- Kirk, P. L.** See Dunn, R.
- Kirkby, W.** Review of Cochrane's *Lavoisier*, 320.
- *Kitto, W. H.** Determination of fat by the Grossfeld method, 97.
- Klentsch, K.** See Täufel, K.
- Knallinsky, A.** See Gatti, C.
- Knock, G. G.** See Baumgartner, J. G.
- *Kny-Jones, F. G., Lindsey, A. J., and Penney, A. C.** The quantitative electro-deposition of tin from chloride solutions. I. Stannic tin solutions, 498.
- Koch, J.** See Diemair, W.
- Kocsis, E. A.** Spot tests for silver, lead, mercury, uranium and aluminium, 190.
- Kodicek, E.** Estimation of nicotinic acid in animal tissues, blood and certain foodstuffs, 522.
- Koehn, C. J., and Sherman, W. C.** Estimation of vitamin A and carotene with the photo-electric colorimeter, 303.
- Koenig, E. W.** Direct determination of alumina in silicates, 180.
- Kolb, J. J., and Toennies, G.** Colour reaction of methionine, 114.
- Kolthoff, I. M., and Langer, A.** Titration of nickel with dimethylglyoxime with the aid of the dropping mercury electrode as indicator electrode, 476.
- Komm, E., and Flügel, J.** Aromatic principle in bread. III. Colorimetric estimation of diacetyl, acetoin and butylene glycol, 361.
- Komm, E., and Lehmann, G.** Aromatic principle in bread. II. Isolation of the aromatic principle in rye bread, 360.
- Komm, E., and Pinder, H.** Determination of creatinine with *m*-dinitrobenzoic acid, 229.
- Kon, G. A. R.** See Whiteley, M. A.
- Kon, S. K.** See Henry, K. M.
- Königstein, M.** See Britton, H. T. S.
- Koolhaas, D. R.** See Meijer, T. M.
- See also van Hulssen, C. J.
- Koonen, H. F.** See Scudi, J. V.
- Kopnaris, G. P.** See Thomis, G. P.
- Koschara, W.** Urothione, a yellow sulphur-containing pigment from human urine, 367.
- Koschara, W., von der Seipen, S., and Altfred, P. A.** Fluorescence analysis of human urine, 242.
- Kouznecov, V. J.** Detection and determination of germanium with hydrogen selenide, 68.
- Koyanagi, H.** See Tsujimoto, M.
- Kramer, B.** See Sobel, A. E.
- Krebs, H. A., Smyth, D. H., and Evans, E. A.** Determination of fumarate and malate in animal tissues, 571.
- Kringstad, H.** See Lunde, G.
- Krishna, S.** See Puntambeka, S. V.
- Kroupa, E.** Micro-gravimetric separation of zinc and uranium, 435.
- Kroupa, F.** See Bisanz, R.
- Krüger, A.** Volumetric determination of sulphate, 434.
- Krussse, A.** See Zwikker, J. J. L.
- Kuhn, P.** See György, P.
- Kurtz, F. E., and Schaffer, P. S.** Separation of hydroxy from non-hydroxy fatty acids by means of a dibasic acid anhydride, 528.

L

- LaForge, F. B., and Acree, F., Jr.** Behaviour of certain substituted allenes towards the Meinel colour test, 614.
- *Lambie, D. A., and Schoeller, W. R.** The separation of molybdenum from tin and from sulphur, 281.
- Lampert, L. M.** Freezing-point of milk, 108.
- Lampitt, L. H.** See Sylvester, N. D.
- Lamprey, H., Sommer, E. E., and Kiffer, A. D.** Detection and determination of ethylene glycol in lubricating oil, 665.
- Landis, Q.** Polariscope determination of proteolytic activity, 660.
- Lang, O. St. A. K., and Adickes, F.** On the fatty acid dehydrogenase IV. product of the dehydrogenation of stearic acid, 241.
- Lang, O. St. A. K., and Mayer, H.** Fatty acid dehydrogenase and its co-enzymes, 241.
- Langer, A.** Amperometric titration of fluoride with thorium nitrate, 669.
- See also Kolthoff, I. M.
- Lantz, L. A.** Standard method for testing the fastness to light of coloured textile materials, 582.
- Lasocka, D.** See Mystkowski, E. M.
- Lederer, R.** See György, P.
- Lees, A.** Appointed Deputy Public Analyst for the Borough of Darwen, 411.
- *Lees, H.** See Grigeman, N. T.
- Lehmann, G.** See Komm, E.
- Lemos, A.** See Cheramy, P.
- Lemos, A. C.** Toxicology of manganese, 63.
- Leong, P. C.** Effect of soil treatment on the vitamin B₁ content of wheat and barley, 59.
- Leopoldi, G.** See Fischer, H.
- Letonoff, T. V., and Reinhold, J. G.** Colorimetric determination of lead as chromate with the aid of diphenylcarbazide, 433.

- Leva, E.** Colorimetric micro-method for the estimation of sodium with manganous uranyl acetate, 316.
- Levine, M., Carpenter, P., and Coblentz, J. M.** Bacteria from chlorinated waters, 370.
- Levy, G. A.** Estimation of barbiturates in blood, 471.
- Levy, L. F.** Identification and determination of phenolic compounds, 430.
- *Lewis, D. T.** The gravimetric determination of phosphate and vanadate, 560.
- Lewis, D. T., and Davis, V. E.** Determination of arsenic in organic and inorganic compounds, 531.
- Lewis, H. B.** See Schultz, J.
- Lewis, J. B.** Determination of sulphide sulphur in presence of other sulphur compounds, 669.
- Lewis, J. B., and Bradstreet, R. B.** Determination of unsaturation in aliphatic hydrocarbon mixtures by bromine absorption, 576.
- Liebhafsky, H. A., and Sharkey, W. H.** Determination of organic peroxides, 475.
- *Lindsey, A. J.** See Kny-Jones, F. G.
- *Lindsey, A. J., and Shennan, R. J.** Quinaldinic acid as a reagent for the separation of copper and cadmium, 636.
- Link, K. P.** See Moore, S.
- *Lipcomb, A. G.** The reaction between 2·2'-dichlorodiethyl sulphide (mustard gas) and bleaching powder, 100.
- Lochte, H. L.** See Schultze, H. G.
- *Lockwood, H. C.** An adjustable support for crucibles and dishes, 160.
- Technique for the preparation of standard microscope slide, with particular reference to chocolate, 97.
- Loeb, H. G.** See Kass, J. P.
- Lollar, R. M.** Determination of chromium in chrome-tanned leather, 531.
- Lotichius, J.** Colorimetric estimation of indigo, 668.
- Colorimetric estimation of sodium hydro-sulphite and the hydrosulphite content of indigo vats, 668.
- Estimation of indigo on the fibre, 668.
- Lovern, J. A., and Morton, R. A.** Distribution of the fatty acids in halibut intestinal oil, with a note on the presence of free fatty acids in the intestines of fish, 58.
- Lovett-Janison, P. L., and Nelson, J. M.** Ascorbic acid oxidase from summer crook-neck squash (*C. pepo condensata*), 572.
- Lucas, A.** Review of Mond and Myers' *Temples of Aramant*, 672.
- Ludeman, C. G.** Determination of sludge in used or oxidised motor oils, 664.
- Lunde, G., and Kringstad, H.** Studies on the anti-grey hair vitamin for silver foxes, 368.
- Lynch, G. Roche, and Pryoe, D. M.** Toxicology: Homicidal, Suicidal and Accidental Poisoning (Review), 139.
- Lynch, H. J., and Deahl, N.** Determination of vanillin in vanilla extracts, 570.
- Lynen, F.** Respiration of animal tissue after freezing in liquid air, 425.
- Lyons, C. G.** See Appleyard, F. N.
- Lythgoe, H. C.** Report of the Director of the Division of Food and Drugs Dept. of Public Health, Commonwealth of Massachusetts, 226.
- *Macara, T.** See Hinton, C. L.
- Macarovich, C. G.** See Spacu, G.
- McCance, R. A.** See Masters, M.
- McCance, R. A., and Widdowson, E. M.** The chemical composition of foods (Medical Research Council Special Report Series, No. 235), 458.
- McCarthy, T. E.** See Sullivan, M. X.
- McCulloch, E. C.** See St. John, J. L.
- MacDonald, K.** Efficiency of the Wells air centrifuge as determined by air-washing technique, 470.
- McIntosh, I. G.** See Grimmett, R. E. R.
- McIntyre, G. H., and Bozain, M.** Optical method for the determination of quartz particles in feldspar, 535.
- MacIntyre, W. H.** See Hammond, J. W.
- McKay, R. W.** Estimation of viscose rayon, 432.
- McKee, R. W.** See Binkley, S. B.
- Mackinney, G.** See West, C. A.
- MacLachlan, E. A.** See Talbot, N. B.
- Macmorran, G. H.** Bleach ointment, 423.
- McNabb, W. M., and Helman, S.** Determination of free cyanide in brass electroplating baths, 315.
- MacNalty, Sir A.** Report of Chief Medical Officer to Ministry of Health for 1938, 103.
- Macrae, T. F.** See El Sadr, M. M.
- Maddison, L.** See Hilditch, T. P.
- *Maddocks, J. R.** See Kersey, R. W.
- Mahr, C., and Ohle, H.** Volumetric determination of chlorate and bromate, 189.
- Volumetric determination of manganese and lead dioxides, 433.
- Mahr, C., and Proding, W.** Non-precipitation of cobalt and palladium by nitro- β -naphthol, 187.
- Maiden, A. M.** See Ford, W. P.
- Maitra, M. K., and Moore, T.** Vitamin A content of "light white" casein, 116.
- Makris, C. G., and Menaché, R.** Colorimetric determination of silver, 477.
- Malaprade, L.** New method for the volumetric determination of bismuth, 249.
- Mallinder, R.** Appointed Agricultural Analyst for the County Boroughs of Burton-on-Trent, Halifax and Huddersfield, 411.
- Appointed Public Analyst for the County Boroughs of Burton-upon-Trent, Halifax and Huddersfield, and the Borough of Scarborough, 411.
- *—** See also Sutton, R. W.
- Malloch, J. G.** Selenium in Canadian wheat, 430.
- Mallya, M. V.** See Vidyarthi, N. L.
- Malowan, L. S.** Detection of gold by means of morpholin, 310.
- Man, E. B.** See Riggs, D. S.
- *Manley, C. H.** The analytical constants of spirit vinegar, 594.
- The estimation of lead in drinking water, 403.
- Mann, F. G., and Saunders, B. C.** Introduction to Practical Organic Chemistry (Review), 74.
- Manning, E. P. D.** See Almquist, H. J.
- Manunta, C.** Chromatographic separation of palmitic and stearic acids from a mixture of oleic, palmitic and stearic acids, 64.

- Marcilla, R.** The Bellier test and the Blarez test. Application to olive and other oils, 170.
- Marensi, A. D., and Banfi, R. F.** Estimation of small amounts of sulphur as sulphates in biological fluids, 366.
- Marloth, B. W.** See Holm, J. M.
- Marsden, A. W.** New cell for electro dialysis, especially of soils, 435.
- Mason, R. D.** Fat, Total Solids and Moisture (Review), 75.
- Masters, M., and McCance, R. A.** Sulphur content of foods, 42.
- Matagrín, A.** Manuel du Savonnier (Review), 389.
- Matthews, J. W.** Review of Conway's *Micro-diffusion Analysis and Volumetric Error*, 254.
- Mattill, H. A.** See Golumbic, C.
- Maudsley, F.** Appointed Public Analyst for the Borough of Colne, 161.
- Mayer, H.** See Lang, O. St. A. K.
- Maynard, L. A.** See Voris, L.
- Masumdar, A.** See Basu, U. P.
- Meade, E. M.** New colour reaction for diaryl-amines, 530.
- Meara, M. L.** See Atherton, D.
- Meara, M. L., and Zaky, Y. A. H.** Fatty acids and glycerides of the seed fats of *Allanblackia floribunda* and *Allanblackia parviflora*, 517.
- Mecchi, E.** See Almqvist, H. J.
- Mehlig, J. P., and Johnson, K. R.** Detection of magnesium by means of *p*-nitrobenzeneazoresorcinol, 314.
- Meijer, T. M.** Essential oil of massoi bark, 375.
- Meijer, T. M., and Koolhaas, D. R.** Determination of rotenone in derris root, 526.
- New derivatives of constituents of derris root, 244.
- Mellanby, E.** See Harrison, D. C.
- Mellon, M. G.** See Howe, D. E.
- Mellor, J. W.** See Parkes, G. D.
- Melnick, D., and Field, H.** Chemical estimation of nicotinic acid: Inhibitory effect of cyanogen bromide upon the aniline side reactions, 661.
- Chemical estimation, stability and form of aneurin in urine, 56.
- Estimation of nicotinic acid in biological materials by means of photoelectric colorimetry, 571.
- Meloche, V. W.** See Hiskey, C. F.
- Ménaché, R.** See Makris, C. G.
- Mendel, H., and Coops, J.** Gradual decomposition by oxidation of fatty acids into their next lower homologues, 246.
- Merrill, H. B., and Mueller, G. O.** Use of cheap filter paper for analysing tan liquors, 476.
- Mertz, J. C.** Determination of chromic oxide in leather. I. Comparison of wet and dry oxidation methods, 310.
- Meyrowitz, R.** See Hart, D.
- Mikschik, E.** See Janke, A.
- Miller, C. C.** Volumetric determination of small quantities of barium and sulphate with barium rhodizonate as indicator, 476.
- Miller, W. T., and Williams, K. T.** Minimum lethal dose of selenium as sodium selenite for horses, mules, cattle and swine, 613.
- Mills, W. H.** Obituary of Sir W. J. Pope, 258.
- *Mitchell, C. A.** An ancient Egyptian marking ink, 100.
- *— Copper in tomato pulp, 220.
- Review of *Chambers's Technical Dictionary*, 622.
- Review of Clark's *Photography by Infra-red: Its Principles and Applications*, 480.
- Review of *The Mineral Water Trade Year Book*, 142.
- Mitchell, W.** New solanaceous alkaloids from *Duboisia myoporoides*, 233, 422.
- Moir, D. D.** Appointed Deputy Public Analyst for the Urban District of Esher and the Borough of Surbiton, 27; for the Urban Districts of Coulsdon and Purley, and Merton and Morden, and the Borough of Wimbledon, 161; for the Urban District of Carshalton, and the Borough of Mitcham, 411.
- *— Bleach ointments, 154.
- Moir, G. M., and Andrews, E. D.** Determination of iron and copper in butter, 657.
- Moller, H.** See Keller, H.
- Molnar, N. M., and Grumer, J.** Volumetric estimation of lac in glazed candies, 175.
- Moltano, C. J., and Rapson, W. S.** Seasonal variations in the vitamin A content of certain visceral organs of the Geelbek or Cape salmon (*Atractoscion acqoides* C. and V.), 59.
- Mond, Sir R., and Myers, O. H.** Temples of Armant (Review), 672.
- Monk, H. E.** Appointed Public Analyst for the Borough of Oldbury, 27.
- Moore, F. J.** A History of Chemistry, 3rd Ed. Revised by W. T. Hall and Edited by J. F. Norris (Review), 386.
- Moore, H.** Glasses: Organic and inorganic, 126.
- Moore, M. L.** See Fernholz, E.
- Moore, S., and Link, K. P.** Carbohydrate characterisation. I. Oxidation of aldoses by hypiodite in methanol. II. Identification of seven aldo-monosaccharides as benzimidazole derivatives, 419.
- Moore, T.** See Davies, A. W.
- See also Maitra, M. K.
- Moorhead, F. F.** Determination of carbon disulphide in air, 525.
- Moran, T.** See Brooks, J.
- More, A.** Obituary of W. T. Burgess, 2.
- Morgan, Sir G. T.** Foreword to *Kingszett's Chemical Encyclopaedia*, 6th Ed., 537.
- Morrell, E. S., and Phillips, E. O.** Oxidation of drying oils and cognate substances. VI. Properties of the peroxide, ketol and oxido groups, including those of some resins, 615.
- Morrison, G. K.** Application of rational analysis to some typical South African coals, 376.
- *Morton, R. A.** The constitution and physiological significance of carotene and allied pigments, 263.
- See also Lovern, J. A.
- Motzoc, M. D.** See Balanescu, G.
- Mosolowski, W.** Quantitative estimation of glycuronic acid and its conjugated compounds by means of the naphthoresorcinol test of Tollens, 519.
- Mueller, G. C.** See Merrill, H. B.
- Mukherjee, A.** See Qudrat-i-Khuda, M.
- Müller, E., and Burchard, C.** New method for the removal of nitric acid, 380.

- Müller, E., and Stumpf, W. Determination of iodide in mineral waters containing bromide and large quantities of chloride, 245.
 Müller, G. See Geiger, E.
 *Murti, K. S. See Hilditch, T. P.
 Murty, G. N. Report of Public Analyst for Municipality of Bangalore for 1939-40, 353.
 Muter, A. H. M. Appointed Public Analyst for the Metropolitan Borough of Battersea, 161.
 Myers, O. H. See Mond, Sir R.
 Mystkowski, E. M., and Lasocka, D. Factors preventing oxidation of ascorbic acid in blood serum, 59.

N

- Naidu, S. R. Report of Chemical Examiner for Government of Madras for 1938, 36; for 1939, 602.
 Nakamura, T. Determination of silicon in fluorspar and cryolite, 434.
 Nakamura, T., and Yamasaki, S. Determination of aluminium oxide in metallic aluminium by means of cupric ammonium chloride, 68.
 Nance, J. T. See Guthrie, F. C.
 Narasimhamurthy, G., and Suryanarayanamurthy, V. V. Detection of adulteration in ghee by a study of its fluorescence, 606.
 Neale, S. M., and Stringfellow, W. A. Absorption of substantive dyes by oxycelluloses of the acidic type, 247.
 Nelson, C. I., and Berrigan, D. Effectiveness of heat penetration in the canning of meat in the home by the pressure cooker, 120.
 Nelson, E. K., and Wheeler, D. H. Natural ageing of wines, 43.
 Nelson, J. M. See Lovett-Janison, P. L.
 Nemeo, V. Precipitation of natural tannins with calcium hydroxide, 246.
 *Neubauer, L. G. See Andrew, R. L.
 Neugebauer, H. See Brunner, K.
 Newton, A., and Buckler, E. J. Determination of methylpropene by means of a modified Deniges reagent, 472.
 Nicholls, J. R. Review of Radley and Grant's *Fluorescence Analysis in Ultra-Violet Light*, 3rd Ed., 194.
 Nicholls, J. R. Review of Reilly and Rae's *Physico-Chemical Methods*, 3rd Ed., 384.
 Nicholls, J. R. Review of *The British Encyclopaedia of Medical Practice*, Vol. 12, 139.
 Nickelson, M. N., and Werkman, C. H. Formation of trimethylene glycol from glycerol by *Aerobacter*, 613.
 Nicolas, Y. See Guillaume, A.
 Nielsen, J. P. Determination of magnesium in biological material, 176.
 Nihlen, E. K. See Graff, J. H.
 Nobori, H. Composition of the fatty acids in Chinese coconut oil, 658.
 — Lower saturated fatty acids in herring oil, 607.
 Noetzel, O. Estimation of silver in catadynised vinegar and fruit cordials, 364.
 Norris, F. A. See Kass, J. P.
 Norris, J. F. Editor of Moore's *A History of Chemistry*, 3rd Ed., 386.
 Norris, L. C. See Hodson, A. Z.
 Norton, A. R. See Seaman, W.

O

- O'Dwyer, M. H. Hemicelluloses of the wood of English oak. 5. Structure of hemicellulose, 474.
 Ohle, H. See Mahr, C.
 Okell, F. L. Review of Cummin and Kay's *A Text-book of Quantitative Chemical Analysis*, 7th Ed., 138.
 — Review of Vogel's *A Text-book of Quantitative Inorganic Analysis*, 195.
 — Review of Yoe's *A Laboratory Manual of Qualitative Analysis*, 254.
 Olliver, M. Antiscorbutic values of fruits and vegetables, 662.
 Orchard, E. R. Compost: its preparation and use, 416.

P

- Page, J. See Brooks, J.
 Page, R. P. Appointed Public Analyst for the Borough of Gosport, 411.
 Pal, J. C., and Guha, B. C. Combined ascorbic acid in plant foodstuffs. I. 522.
 Panouse-Digeaud, L., and Oheffel, H. Elimination of iron by cupferron prior to colorimetric determination of lead with dithizone, 130.
 Papavassilou, M. See Cheramy, P.
 Paranjothy, J. T. Rat virus enquiry report (F. M. S. Institute for Medical Research Bull. No. 1, 1939), 460.
 Park, B. Partial separation of copper from small amounts of arsenic, 378.
 Parkes, G. D., and Mellor, J. W. Mellor's Modern Inorganic Chemistry, 9th Ed. (Review), 479.
 Parpart, A. K. See Taylor, A. R.
 Parrish, W. See Kaiser, E. P.
 Paulson, R. A. See Benedetti-Pichler, A. A.
 Payfer, R. See Grossfeld, J.
 Peacock, G. See Emmett, A. D.
 Pearson, Miss M. H. Appointed Deputy Public Analyst for the County of Middlesex, 27.
 Pedelty, W. H. See Hilditch, T. P.
 Pell-Walpole, W. T. Determination of tellurium in tin-rich alloys by volatilisation, 434.
 *Penney, A. C. See Kny-Jones, F. G.
 Pennington, D., Snell, E. E., and Williams, R. J. Assay method for pantothenic acid, 661.
 Perlstein, D. See Gershenfeld, L.
 Péronnet, M., and Guenin, J. Action of a cuprous iodide reagent on alkaloids: precipitation and colour reactions, 301.
 Péronnet, M., and Rémy, R. H. Precipitation reactions of organic arsenic compounds, 247.
 Pesetz, M. New colour reaction for the identification of cocaine, 48.
 Peters, D. D. See Sobel, A. E.
 Phillips, E. O. See Morrell, R. S.
 *Phillips, L. See Britton, H. T. S.
 Phillips, M., and Goss, M. J. Composition of certain nutshells, 608.
 Pinder, H. See Komm, E.
 *Pinder, J. L. The routine examination of magnesium trisilicate, 90.
 *Pinder, J. L., and Singer, J. H. Examination and determination of 2-methyl-1:4-naphthoquinone, 7.

- Pirtea, T. I.** Gravimetric determination of antimony by means of 8-hydroxyquinoline, 188.
- Pittman, F. K.** Detection and elimination of phosphate in qualitative analysis by means of zirconium salts, 669.
- Plank, J.** Estimation of cerium in cerous salts, 434.
- Plimmer, V. G.** Food Values at a Glance (Review), 388.
- Polacek-Wittek, A.** See Schmid, L.
- Popesco, C.** See Ionesco-Matiu, A.
- *Prideaux, E. B. R., and Hennes, J. R.** The precipitation of aluminium hydroxide and its solubility in ammonia, 83.
- Prodinger, W.** See Mahr, C.
- *Proffitt, P. M. C.** See Chirnside, R. C.
- Prudhomme, R. O.** Colorimetric estimation of quinine in biological fluids and tissues, 239.
- Pryce, D. M.** See Lynch, G. Roche.
- *Puffeles, M.** Observations on the use of electrodialysis in the analysis of soils, 501.
- Pulley, G. N., and von Loesecke, H. W.** Gases in the commercial handling of citrus juices, 42.
- Puntambaka, S. V., and Krishna, S.** Fatty oil from the seeds of *Bauhinia variegata*, 421.
- Pyke, M.** Distribution of aneurin in foods determined by chemical analysis, 180.
- Observations of the distribution of vitamin B₁ in some plant families, 470.
- Review of Plimmer's *Food Values at a Glance*, 388.
- Review of Symposium of the Food Group of the Society of Chemical Industry on Vitamin E, 77.
- Q**
- Qudrat-I-Khuda, M., Ghosh, S. K., and Mukherjee, A.** Neem oil. I. 465.
- R**
- Rabak, W.** Factors affecting the efficiency of materials used in packaging frozen foods, 658.
- Rabaté, J.** See Charaux, C.
- Racker, E.** Histidine detection and estimation in urine, 467.
- Radley, J. A.** Starch and Its Derivatives (Review), 481.
- Radley, J. A., and Grant, J.** Fluorescence Analysis in Ultra-Violet Light, 3rd Ed. (Review), 194.
- Rae, R.** See Robinson, R. A.
- Rae, W. N.** See Reilly, J.
- Raistrick, H.** See Anslow, W. K.
- Rapson, W. S.** See Molteno, C. J.
- Ray, P. R., and Sarkar, T. C.** Quinaldinic acid as a micro-reagent. IV. Determination of zinc in presence of copper, silver and mercury, 435.
- Räy, T. W.** Estimation of manganese in organic material containing large amounts of calcium and chlorides, 570.
- Raymond, W. D.** See Harris, L. J.
- Rees, M. W.** See Chibnall, A. C.
- Reilly, J., and Rae, W. N.** Physico-Chemical Methods, 3rd Ed. (Review), 384.
- Reimers, F.** See Jackerott, K. A.
- Reindollar, W. F.** Detection of saccharin in foodstuffs, 569.
- Reinhold, J. G.** See Letonoff, T. V.
- Rémy, R. H.** See Péronnet, M.
- Revukas, A. J.** See Evans, R. N.
- Reynolds, O. V.** Appointed Deputy Public Analyst for the Borough of Torquay, 161.
- Rhodes, H. T. F.** Forensic Chemistry (Review), 621.
- Riemenschneider, R. W.** See Wheeler, D. H.
- Rigby, W. T.** Appointed Public Analyst for the Borough of Rugby, 27.
- Riggs, D. S., and Man, E. B.** Permanganate acid ashing micro-method for iodine determinations. 1. Values in blood of normal subjects, 579.
- Rijks-Instituut Voor Pharmaco-Therapeutisch Onderzoek.** Contraphthisine, 48.
- Ripan-Tilici, R.** Argentometric determination of cyanide with diphenylcarbazone as indicator, 315.
- Roberts, E. A. H.** Fermentation process in tea manufacture. V. Cytochrome oxidase and its probable rôle. VI. Effect of dilution on the rate and extent of oxidations in fermenting tea leaf suspensions, 521.
- See also Harrison, C. J.
- Roberts, R. F.** See Haslam, J.
- Roberts, W. H.** Appointed Public Analyst for the Boroughs of Crosby and Kendal, 411.
- Robertshaw, G. F.** See Burton, D.
- Robertson, J. D.** Urine Examination (Review), 139.
- Robinson, C. H., and Sykes, W. J.** Obituary of F. T. Shutt, 392.
- Robinson, F. A.** Review of Anderson's *Essentials of Physiological Chemistry*, 76.
- Review of Appleyard and Lyons' *Practical Pharmaceutical Chemistry*, 4th Ed., 436.
- Review of Kingzett's *Chemical Encyclopaedia*, 6th Ed., 537.
- Robinson, H. W., and Hogden, C. G.** Biuret reaction in the estimation of serum proteins. I. Study of the conditions necessary for the production of a stable colour which bears a quantitative relationship to the protein concentration, 466.
- Robinson, R. A., and Rae, R.** Bell's Sale of Food and Drugs, 10th Ed. (Review), 385.
- *Robinson, R. G.** A simple test for arsenic in lead alloys, 159.
- Rochow, E. G.** Preparation of Grignard reagents from magnesium amalgams, 245.
- Roelen, L.** Colorimetric determination of iron in aluminium and alumina, 187.
- Rogers, W. J., Beamish, F. E., and Russell, D. S.** Determination of ruthenium with thionalide, 669.
- Rosenthaler, L.** Detection of elements in organic compounds, 307.
- Rowan, P. A., and Insinger, J. A.** Determination of the eugenol-content of essential oils, 66.
- Russell, D. S.** See Rogers, W. J.
- Russell, E. W., and Tamhane, R. V.** Determination of the size distribution of soil clods and crumbs, 371.

- Saber, A. H.** Quantitative determination of powdered cinnamon and cassia, 421.
- Sabetay, S.** Phthalisation in the presence of pyridine, 125.
- Sachanen, A. N.** Conversion of Petroleum (Review), 671.
- Sadolin, E.** Analysis of synthetic resins containing maleic acid, 128.
- Sage, C. E.** Review of Moore's *A History of Chemistry*, 3rd Ed., 387.
- Sah, P. P. T.** 3,5-Dinitro-4-methylbenzazide as a reagent for the identification of amines, 125.
— *p*-Nitrobenzazide and *p*-nitrophenyl isocyanate as reagents for the identification of amines, 374.
- Sah, P. P. T., and Woo, T. F.** *m*-Nitrobenzazide as a reagent for the identification of phenols, 124.
- Sah, P. P. T., and Yin, W.-H.** *o*-Nitrobenzazide as a reagent for the identification of phenols, 373.
- St. John, J. L., and Others.** Toxicity to sheep of lead arsenate and lead arsenate spray residues, 575.
- Samson, M.** Relation of cuprous creatinine to tests for sugar in urine, 55.
- Sand, H. J. S.** Electrochemistry and Electrochemical Analysis. Vol. II. Gravimetric Electrolytic Analysis and Electrolytic Marsh Tests (Review), 620.
- Sandhoff, A. G.** See Fuchs, W.
- *Sandilands, J.** The assay of mercury, 13.
- Sandler, L.** See Bergmann, E.
- Sando, C. E.** See Wheeler, D. H.
- Sarkar, T. C.** See Rây, P. R.
- Saul, S.** See Hilditch, T. P.
- Saunders, B. C.** See Mann, F. G.
- Scarborough, H.** Vitamin P, 61.
- Schaffer, P. S.** See Kurtz, F. E.
- Schlitter, E.** See Hartmann, H.
- Schlösser, M. A.** See Graff, J. H.
- Schmid, L., and Polaczek-Witte, A.** Colouring matters of flowers of *Tormentilla potentilla*, 309.
- Schoch, T. J., and Jensen, C. C.** Simplified alkali-lability determination for starch products, 655.
- Schoder, W. P.** See Weinig, A. J.
- Schoeller, W. R.** Review of Weinig and Schoder's *Technical Methods of Ore Analysis for Chemists and Colleges*, 11th Ed., 194.
*— See also Lambie, D. A.
- Schoenheimer, R.** See Bernhardt, K.
- Scholten, C.** Estimation of carbon monoxide in blood, 471.
- Schoorl, N.** Little-known reaction for benzoic acid, 374.
- *Schormüller, J.** Ultra-violet absorption of yeast extracts and meat extracts, 297.
- Schou, S. A., and Bennakou, J.** Colorimetric determination of ergotoxine and ergometrine, 51.
- Schulte, K. E.** See Thaler, H.
- Schulte, M. J.** Reactions for the identification of isazin, 112.
- Schultz, J., and Lewis, H. B.** Excretion of volatile selenium compounds after administration of sodium selenite to white rats, 302.
- Schultze, H. G., Shive, B., and Lochte, H. L.** Separation and characterisation of petroleum acids, 529.
- Schwarz, G.** Photographic silver-gelatin paper as a reagent in spot analysis, 619.
- Schweitzer, T. E.** See Hoffman, C.
- Scott, D. B.** Detection of the bleaching of flour with chlorine, 605.
- Scott, E. W., and Treon, J. F.** Colorimetric determination of primary mononitroparaffins, 529.
- Soudi, J. V., Koonas, H. F., and Keresztesy, J. C.** Estimation of adermin (vitamin B₆) in urine, 304.
- *Seaber, W. M.** Alumina for the estimation of carotene, 645.
*— The commercial determination of carotene and allied pigments, with especial reference to dried grass and other leafy materials, 266.
- Seaman, W., Norton, A. E., and Sundberg, O. E.** Estimation of *o*-nitrophenol in *p*-nitrophenol, and *o*-aminophenol in *p*-aminophenol by fluorescence and analysis, 581.
- Seath, J., and Beamish, F. E.** Assay for platinum metals in ore concentrates, 310.
- Seikel, M. K.** Identification of aromatic halogenonitro compounds as piperidyl derivatives, 666.
— Identification of organic compounds. II. Piperidyl derivatives of aromatic halogenonitro compounds, 530.
- +Sen-Gupta, P. N., and Guha, B. C.** Combined ascorbic acid in plant foodstuffs. II, 523.
+ Estimation of vitamin C in foodstuffs, 523.
- Serrure, G.** See Gaspard, R.
- Servigne, M.** Luminescence analysis of rare elements, 382.
- Shannon, W. J., and Englis, D. T.** Copper in tomatoes, 605.
- Sharkey, W. H.** See Liebhafsky, H. A.
- Sharp, E. A.** See Emmett, A. D.
- *Shennan, R. J.** See Lindsey, A. J.
- Sherman, C. S., and Herzog, R.** Determination of pyrethrin I. The soil colour reaction in the mercury reduction method, 373.
- Sherman, W. C.** See Koehn, C. J.
- Sherwood, F. W.** See Halverson, J. O.
- Shimwell, J. L.** Classification and identification of bacteria with special reference to the beer types, 612.
- Shinowara, G. Y., and Brown, J. B.** Isolation of arachidonic acid, 576.
- Shive, B.** See Schultze, H. G.
- Shorland, F. B.** New Zealand fish oils. III. Composition of the depot fats of the ling (*Genypterus blacodes*), 172.
- Shrikhande, J. G.** Estimation of lignin in tannin materials, 530.
- Shupe, I. S.** Separation and determination of 4-aminodiphenylamine, 308.
— Separation and determination of 2,4-diaminodiphenylamine, 666.
— Separation of *p*-aminophenol, *p*-methylaminophenol and *o*-aminophenol, 667.
- Sideris, C. P.** Colorimetric determination of magnesium, 478.
— Improved form of the formaldoxime colorimetric method for manganese, 476.
- Silva, M.** Macatiba oil (mocaya), 605.

- ***Simmons, J. E.** A new method for the determination of free acid in sulphate of alumina, alum cake, etc., 456.
- Simmons, W. H.** Review of Matagrin's *Manuel du Savonnier*, 389.
- Simons, E. J., Buxton, L. O., and Colman, H. B.** Vitamin A destruction in fish-liver oils, 469.
- Simpson, E. S.** Report of the Government Analyst for Western Australia on the work of the Chemical Branch, Mines Dept., for 1938, 291.
- Simpson, I. A.** The antineuritic value of par-boiled rice. (F.M.S. Institute of Medical Research Bull. No. 4, 1939), 461.
- Sinclair, H. M.** Estimation of aneurin in blood. II. Further modification of Meiklejohn's method, 182.
- Estimation of vitamin B₁ in cerebro-spinal fluid, 181.
- ***Singer, J. H.** See Pinder, J. L.
- Singh, B. and S.** Iodimetric determination of organic acids, 186.
- Singruen, E.** Oxidation turbidities (in beer) and their causes, 569.
- Sirdar, J. N.** See Bagchi, K. N.
- Sjollem, B., and Donath, W. F.** Vitamin A, carotene and xanthophyll content of the yolk of hens' eggs, 521.
- Sloley, R. W.** Review of Holmstrom's *Records and Research in Engineering and Industrial Science*, 322.
- Slooff, A., and Van Duyn, D.** Determination of chlorides, thiocyanates and cyanides in presence of each other, and a qualitative reaction for the detection of chlorides in potassium thiocyanate, 250.
- Smith, G. F.** See Duke, F. R.
- Smith, G. F., and Getz, C. A.** Cerate oxidimetry. Stability of solutions, 532.
- ***Smith, G. S.** The analysis of cadmium-plating solutions, 87.
- Smith, L. I., Irwin, W. B., and Ungnade, H. E.** Vitamin E chemistry: Oxidation products of α -tocopherol and of related 6-hydroxy-chromans, 115.
- Smyth, D. H.** See Krebs, H. A.
- Snell, E. E.** See Pennington, D.
- Snell, E. E., Eakin, R. E., and Williams, R. J.** Quantitative test for biotin and observations regarding its occurrence and properties, 524.
- Snell, F. D.** See Epstein, S. S.
- Snider, S. R.** Determination of amino-nitrogen in malt extracts, 464.
- Sobel, A. E., Yusha, H., Peters, D. D., and Kramer, B.** Biochemical behaviour of lead. I. Influence of calcium, phosphorus and vitamin D on lead in blood and bone, 235.
- Society of Chemical Industry:** Annual Reports of the Progress of Applied Chemistry. Vol. XXIV for 1939. (Review), 436.
- Sommer, E. E.** See Lamprey, H.
- Sommer, H.** Colour test for elemental sulphur, 532.
- Soroos, H.** Lead-sodium alloy as a drying agent, 191.
- Sotola, J.** See St. John, J. L.
- Spacu, G., and Macaroviici, C. G.** Reaction of copper with benzidine, 248.
- Spalding, R. C.** See Ford, W. P.
- Spiegelhaeck, H.** Determination of silica in cryolite and materials containing fluorine, 315.
- Springer, V. F.** See Hiskey, C. F.
- Spuhr, W. P.** See Sterling, C.
- Stavely, H. E.** See Fernholz, E.
- Steger, A., and van Loon, J.** Oil from the seeds of *Canarium commune* L., 230.
- Stein, W. H.** See Bergmann, M.
- Sterling, C., and Spuhr, W. P.** Determination of molybdenum in cast iron and steel, 311.
- Stevens, R. E.** Modification of the J. Lawrence Smith method for the extraction of alkalis in rocks, 616.
- Stevenson, W. W.** See Swinden, T.
- Stewart, A. W.** Appointed Public Analyst for the Urban District of Thurrock, 411.
- Stillings, R. A., and Browning, B. L.** Photometric estimation of furfural, 663.
- Stock, C. J. H.** Appointed Public Analyst for the Borough of Stockton-on-Tees, 411.
- Stokstad, E. L. R.** See Almquist, H. J.
- Stone, I.** Preparation of stable 2,6-dichlorophenol-indophenol solutions, 660.
- Straub, J., and Van Amerongen, G. J.** Microscopical investigation of true woollen cloths. Chemical and microscopical determination of additions of casein wool, 582.
- Stringfellow, W. A.** See Neale, S. M.
- Strong, R. K.** Revision of *Kingzett's Chemical Encyclopaedia*, 6th Ed. (Review), 537.
- Stubblefield, F. M., and Deturk, E. E.** Effect of ferric sulphate in shortening the digestion period in the Kjeldahl method, 616.
- Stubbs, J. R.** Appointed Public Analyst for the Borough of Leigh (Lancs) and the Urban District of Newton-le-Willows, 27; for the Borough of Widnes, 161; for the Boroughs of Darwen, and of Morecombe and Heysham, 411.
- Stumpt, W.** See Müller, E.
- Sullivan, J. T.** Determination of hydrocyanic acid by the picric acid method and the KWSZ photometer, 122.
- Sullivan, M. X., and McCarthy, T. E.** Colorimetric test for methionine, 466.
- Sumner, J. B. and R. J.** Coupled oxidation of carotene and fat by carotene oxidase, 571.
- Sundara Rao, W. V.** See Gopala Rao, G.
- Sundberg, O. E.** See Seaman, W.
- Suryanarayanamurty, V. V.** See Narasimhamurty, G.
- ***Sutton, R. W., and Hitchen, O.** The determination of benzoic acid, 502.
- ***Sutton, R. W., and Others.** The examination of lard, 623.
- Swinden, T., and Stevenson, W. W.** Accelerated spray test for determining the relative atmospheric corrodibility of ferrous materials, 670.
- Sykes, W. J.** See Robinson, C. H.
- Sylvester, N. D., and Lampitt, L. H.** Determination of cobalt in foods, 424.
- Synge, R. L. M.** Experiments on amino acids, 238.
- Szebelledy, L., and Ajtai, M.** Activation of vanadium with pyrocatechol, 191.
- Szép, Ö.** See Bodnár, J.

T

- Takano, M., Takao, M., and Danjo, M.** Selective action of fatty acid on the ethanolysis of sardine oil, 607.
- Takao, M.** See Takano, M.
- Talbot, N. B., and Others.** Colorimetric assay of weakly phenolic ketones, "oestrone," in extracts of human urine, 609.
- Tamhane, R. V.** See Russell, E. W.
- Tannich, R. E.** See Gilbert, G. R.
- *Tate, F. G. H., and Whalley, H. K.** The spectrographic analysis of tobacco ash, 587.
- Taufel, K., and Klentsch, K.** Chemistry of fat spoilage. Analytical differentiation of the aldehydes formed, 44.
- Taylor, A. R., Parpart, A. K., and Ballentine, R.** A rapid circulating dialyser, 253.
- Taylor, G.** Appointed Joint Public Analyst for the Borough of Ilford, 27; for the Borough of Wanstead and Woodford, 161.
- Taylor, H.** Toxicity of coal tar naphtha distillates, 121.
- Taylor, W. W.** Appointed Additional Public Analyst for the Borough of Kesteven, 27; Public Analyst for the Borough of Mansfield, 411.
- Tchitchibabine, A. E., and Hoffman, Ch.** Identification of plasmoguin by a colour reaction, 47.
- Teller, E.** See Hibben, J. H.
- Thaler, H., and Schulte, K. E.** Detection of artificial dyes in tomato pulp, 300.
- Thayer, S. A.** See Binkley, S. B.
- Thomas, G. N., and Kopanaris, G. P.** Determination of bismuth, iodine and quinine in quinine iodobismuthate and its injectable preparations, 49.
- Thompson, M. de K.** Theoretical and Applied Electrochemistry, 3rd Ed. (Review), 193.
- Thorne, P. C. L., and Ward, A. M.** Translation of Ephraim's *Inorganic Chemistry*, 3rd English Ed., 584.
- Thorp, C. E.** Starch-iodide method for ozone, 525.
- Thorpe, Sir J. F., and Whiteley, M. A.** Thorpe's Dictionary of Applied Chemistry, 4th Ed. (Review), 583.
- Thuret, J.** Permanent artificial standards for the nephelometric determination of arsenic with Bougault's reagent, 129.
- Tickle, T.** Appointed Public Analyst for the Borough of Torquay, 161.
- Tincker, M. A. H., and Unwin, C. H.** Root-forming substances used for propagation purposes, 119.
- Tocher, J. F.** Report of the Public Analyst for the County of Aberdeen for 1938, 28.
- Todd, A. R.** See Jacob, A.
- Todhunter, E. N.** See St. John, J. L.
- Toennies, G.** See Kolb, J. J.
- Toms, H.** Review of Engelder's *Calculations of Quantitative Analysis*, 674.
- Review of Gordon and Trout's *Introductory College Chemistry*, 2nd Ed., 537.
- Review of Mann and Saunders' *Introduction to Practical Organic Chemistry*, 74.
- Török, T.** Spectrovolumetric determination of alkaline earths and phosphate, 383.
- See also Bodnár, J.
- Townsend, F. E., and Gade, G. N.** Separation of cadmium from zinc by precipitation with aluminium, 310.
- Treon, J. F.** See Scott, E. W.
- Trout, W. E., Jr.** See Gordon, N.
- Truog, E.** See Berger, K. C.
- See also Bower, C. A.
- Trusler, E. B.** Effect of glycerin on the distillation method for water, 665.
- Tsujimoto, M.** Bamboo leaf wax, 187.
- Linolenic acid in the leaves of land plants, 664.
- Tsujimoto, M., and Koyanagi, H.** Liver oils of some terrestrial animals, 231.
- Tsurugi, J.** See Kimura, W.
- Tweeney, C. F., and Hughes, L. E. C.** Editors of *Chambers's Technical Dictionary* (Review), 622.

U

- Ungnade, H. E.** See Smith, L. I.
- Unwin, C. H.** See Tincker, M. A. H.
- Urech, P.** Colorimetric estimation of silica and its use in the metallurgy of aluminium, 131.
- Vallance, R. H.** See Friend, J. N.
- Van Amerongen, G. J.** See Straub, J.

V

- Van Der Meulen, J. H.** Analysis of chrome green and similarly prepared pigments, 133.
- Van Duyn, D.** See Slooff, A.
- van Hulssen, O. J., and Koolhaas, D. R.** Determination of catechin and catechin-tans in gambir, 127.
- van Loon, J.** See Steger, A.
- Van Pinxteren, J. A. C.** Alkaloid-content of *Extractum hyoscyami*, 172.
- van Zyl, J. P.** Report of the Division of Chemical Services, South Africa, for 1939, 229.
- Verda, A., and Herzfeld, E.** Rapid method for the determination of nicotine, 234.
- Vesterhus, R.** See Aschehoug, E.
- Vickery, H. B.** Estimation of arginine by means of flavianic acid, 236.
- Vidyardhi, N. L., and Dasa Rao, C. J.** Fatty acids and glycerides of the fat from the seeds of *Garcinia indica* (kokum butter), 171.
- Vidyardhi, N. L., and Mallia, M. V.** Component glycerides of vegetable fatty oils. Niger-seed oil, 419.
- Fatty acids and glycerides of the oil from sapota seeds, 171.
- Occurrence of an isomer of ricinoleic acid in the fatty oil from the seeds of *Vernonia anthelmintica*, 186.
- Viennet, E. M.** See Ayres, G. H.
- Voelcker, E.** Appointed Agricultural Chemist for the County Council of the West Riding of Yorkshire, 411.
- Report of the Consulting Chemist to the Royal Agricultural Society for 1939, 462.
- Review of Robinson and Ives' *Bell's Sale of Food and Drugs*, 10th Ed., 385.
- Vogel, A. I.** A Textbook of Quantitative Inorganic Analysis (Review), 195.

- Volmar, Y., and Wagner, A.** Colorimetric determination of ferric iron by means of gallic acid, 249.
- von der Seipen, S.** See Koschara, W.
- von Loesecke, H. W.** See Pulley, G. N.
- Voris, L., Ellis, G., and Maynard, L. A.** Determination of neutral fat glycerol in blood with periodate, 425.

W

- Wagenaar, G. H.** Identification of perchlorate, persulphate and certain other inorganic acid radicals with Zwikker's reagent. A sensitive reaction for copper, 533.
- Wagenaar, M.** Application of the herapathite reaction to aristoin, 111.
- Wagner, A.** See Volmar, Y.
- Wagner, K. H.** Experimental avitaminosis A in man, 426.
- Wain, R. L.** See Glasscock, H. H.
- Walker, B. S.** See Barkan, G.
- See also Darnell, M. C., Jr.
- Walker, G. H.** Appointed Public Analyst for the Borough of Eccles, 27; for the Borough of Stretford, 161.
- Report of the Public Analyst for the City of Salford for 1939, 503.
- Walsh, E. O'F.** See Goldberger, W.
- Walter, F. G.** The Manufacture of Compressed Yeast (Review), 586.
- Walton, S. G.** Report of the Government Analyst for New South Wales for 1937, 599; for 1938, 649.
- Wan, S. W., and Hu, D. B.** Reaction of Wijs' solution with tung oil, 64.
- Ward, A. M.** See Thorne, P. C. L.
- Ward, T. J.** Review of Fiene and Blumenthal's *Handbook of Food Manufacture*, 255.
- Review of Lloyd Hind's *Brewing Science and Practice. Vol. II. Brewing Processes*, 482.
- Review of Rhodes's *Forensic Chemistry*, 621.
- Review of Walter's *The Manufacture of Compressed Yeast*, 586.
- Weast, C. A., and Mackinney, G.** Chlorophyllase, 426.
- Weber, J. H.** Appointed Deputy Public Analyst for the Borough of Windsor (New), 161; for the County Borough of Oxford, 566.
- Weeks, M. E.** Discovery of the Elements, 4th Ed. (Review), 321.
- Weinig, A. J., and Schroder, W. P.** Technical Methods of Ore Analysis for Chemists and Colleges, 11th Ed. (Review), 194.
- *Weir, E. M.** See Dickinson, D.
- Weiser, H. B.** Colloid Chemistry (A Textbook). (Review), 536.
- Wenger, P.** See Cimerman, C.
- Werkman, C. H.** See Nickelson, M. N.
- *Werner, A. E. A.** The determination of thiourea by oxidation with selenious acid, 286.
- Wernimont, G., and Hopkinson, F. J.** Micro-titration of selenium, 534.
- *Whalley, H. K.** See Tate, F. G. H.
- Wheeler, D. H.** See Nelson, E. K.
- Wheeler, D. H., Riemenschneider, R. W., and Sando, C. E.** Preparation, properties and thiocyanogen absorption of triolein and trilinolin, 307.
- Wheeler, R. V.** See Belcher, R.
- Wheeler, T. S.** Report of the State Chemist for Eire for 1939-40, 459.
- Whitby, G. S.** Effects of sulphur dioxide on vegetation, 51.
- White, J.** Effect of the oral administration of dimethylaminoazobenzene (butter yellow) on the growth of the rat, 467.
- Whiteley, M. A.** See Thorpe, J. F.
- Whiteley, M. A., and Kon, G. A. R.** Obituary of Sir J. F. Thorpe, 483.
- Whitmore, W. F., and Wood, C. A.** Chemical microscopy of some toxicological alkaloids, 70.
- Wiegand, H.** See Biedebach, F.
- Wilkoff, H. L., Cohen, B. R., and Grossman, M. J.** Low-temperature cooling baths, 383.
- *Wilkinson, H.** See Gridgeman, N. T.
- Williams, E. F.** See Chibnall, A. C.
- Williams, H. A.** Appointed Public Analyst for the Borough of Beddington and Wallington, 411.
- *Williams, K. A.** Detection of beef fat in lard (Bömer number), 596.
- Review of Burton and Robertshaw's *Sulphated Oils and Allied Products*, 318.
- Williams, K. T.** See Miller, W. T.
- Williams, R. J.** See Pennington, D.
- See also Snell, E. E.
- Wilson, C. L.** A gas detection apparatus for qualitative microchemistry, 407.
- Micro-tests for elements in organic compounds. II. Phosphorus, arsenic and antimony, 405.
- Test for iodide and nitrite, 478.
- Wilson, C. W.** Boric acid colour reaction of flavone derivatives, 66.
- Wilson, D. T.** See Hilty, W. W.
- With, T. K.** See Jensen, H. B.
- Wittmeyer, H.** See Glimm, E.
- Wokes, F.** See Daglish, C.
- Wolfe, J. K.** See Talbot, N. B.
- Woo, T. F.** See Sah, P. P. T.
- Wood, C. A.** See Whitmore, W. F.
- Wood, D. R.** Review of Fairbrother's *Textbook of Bacteriology*, 3rd Ed., 673.
- Woodhead, S. A.** Appointed Public Analyst for the Borough of Worthing, 27.
- Woodmansey, A.** The presence of sulphide in (Harrogate) mineral water and its oxidation by air, 430.
- Work, C. E.** See El Sadr, M. M.
- Wortis, H.** See Bueding, E.
- *Wright, M. D.** See Baker, A. Z.
- Wright, R. F.** Appointed Additional Public Analyst for the Borough of Worthing, 27.

Y

- Yamazaki, S.** See Nakamura, T.
- Yanowski, L. V.** See Hynes, W. A.
- Yanowski, L. V., and Hynes, W. A.** Salts of complex cations applied to the microscopic detection of anions. VIII. 1,2-Chloro-aquotetramminocobaltic chloride, 190.
- Yin, W.-H.** See Sah, P. P. T.
- Yoe, J. H.** A Laboratory Manual of Qualitative Analysis (Review), 254.

Yoe, J. H., and Barton, C. J. Colorimetric determination of copper with triethanolamine, 578.

Young, G. T. *See* Hawkins, E. G. E.

***Young, J. H.** Detection of small amounts of proteins on the surface of metal plates, 564.

***—** Sampling of printed paper, 596.

Yuska, H. *See* Sobel, A. E.

Z

Zondek, S. G. *See* Brückmann, G.

Zwicker, J. J. L., and Kraysse, A. Precipitation of alkaloids with cuprous chloride, 173.

INDEX TO SUBJECTS

* Denotes original papers and notes published in *The Analyst*.

A

Aberdeen: Report of the County Analyst for 1938. J. F. Tocher, 28.

***Absorption Spectra:** Estimation of praseodymium and neodymium in solution from their —. J. N. Friend and D. A. Hall, 144.

Acetamino-acids: Partition of — between immiscible solvents. R. L. M. Synge, 238.

Acetoin: Colorimetric estimation of —. E. Komm and J. Flügel, 361.

Acetone bodies in blood; Method for estimating —. L. A. Crandall, 467.

Determination of alcohol in presence of — in distillates from blood or urine, 600.

Extraction of alkaloids with —. P. Cheramy and M. Papavassilou, 243.

Acetylene in the stimulation of ripening of plums. 225.

N-Acetylhydroxy-amino-acids: Methyl ethers of some —. R. L. M. Synge, 238.

***Acids:** free — in sulphate of alumina, alum cake, etc.; New method of determining. J. R. Simmons, 456.

Acid Radicles: Identification of certain inorganic — with Zwicker's reagent. G. H. Wagenaar, 533.

Acids: Component — of some Indian ox depot fats. T. P. Hilditch and K. S. Murti, 655.
organic —; Iodimetric determination of. B. Singh and S. Singh, 186.

Acridines: Volumetric determination of — with methylene blue. A. Bolliger, 423.

Aderin in urine; Estimation of —. J. V. Scudi, H. F. Koonen and J. C. Keresztesy, 304.

Adulteration in Madras. 601.

Aerobacter: Formation of trimethylene glycol from glycerol by —. M. N. Nickelson and C. H. Werkman, 613.

Agricultural analysis. Abstracts: 122, 184, 244, 305, 371, 430, 526, 574, 613.

Air: aromatic hydrocarbons in —; Detection of. G. R. Gilbert and R. E. Tannich, 666.
carbon disulphide in —; Determination of. F. F. Moorhead, 525.
filters; Testing of —. 599.

*formaldehyde in —; Determination of small amounts of. R. W. Kersey, J. R. Maddocks and T. E. Johnson, 203.

-washing technique; Efficiency of Wells air centrifuge as determined by —. K. MacDonald, 470.

Air Raid Precautions: Lighting restrictions: investigations relating to —. 414.

Albumin: Molecular weight of egg —. F. W. Bernhart, 236.

Alcoholic hydroxyl groups; Rapid qualitative test for —. F. R. Duke and G. F. Smith, 527.

Alcohol in presence of acetone in distillates from blood or urine; Determination of —. 600.

Alcohols: Unsaponifiable matter in fatty —. D. Burton and G. F. Robertshaw, 615.

Aldehydes formed in fat spoilage; Analytical differentiation of —. K. Täufel and K. Klentsch, 44.

Aldo-Monosaccharides: Identification of — as benzimidazole derivatives. S. Moore and K. P. Link, 419.

Aldoses: Oxidation of — by hypiodite in methanol. S. Moore and K. P. Link, 419.

***Alfalfa:** carotene and allied pigments in —; Commercial determination of. W. M. Seaber, 266.

α -Spinasterol isolated from —. E. Fernholz and M. L. Moore, 60.

Aliphatic hydrocarbon mixtures; Determination of unsaturation in — by bromine absorption. J. B. Lewis and R. B. Bradstreet, 576.

Alkali: Iodimetric determination of —. Investigation of G. S. Smith's method. J. Haslam and F. R. Roberts, 399.

Alkaline Earths: Spectroscopic determination of —. T. Török, 383.

Alkalis in rocks; Modification of J. L. Smith method for extracting —. R. E. Stevens, 616.

Alkaloids: Action of cuprous iodide reagent on —. Precipitation and colour reactions. M. Péronnet and J. Guénin, 301.

B.P. Sub-Committee on —. Recommendations. 167.

Chemical microscopy of some toxicological —. W. F. Whitmore and C. A. Wood, 70.

*cocoa — in confectionery; Determination of. J. C. Harral, 408.

content of *Extractum Hyoscyami*. J. A. C. Van Pinxteren, 172.

Extraction of — with acetone. P. Cheramy and M. Papavassilou, 243.

from *Duboisia myoporoides*; New solanaceous —. W. Mitchell, 233.

in preparations of *Berberis vulgaris* and *Berberis agrifolium*; Determination of —. K. Brunner and H. Neugebauer, 47.

Indirect volumetric determination of —. G. Auguste, 110.

of some *Chondrodendron* species, and origin of *Radix paeirae bravas*. H. King, 518.

Poisonous — of *Holarrhena antidysenterica*, Wall. 602.

Precipitation of — with cuprous chloride. J. J. L. Zwicker and A. Kruyse, 173.

Solanaceous — from *Duboisia myoporoides*. W. Mitchell, 422.

Allanblachia Species: Fatty acids and glycerides of seed fats of —. M. L. Meara and Y. A. H. Zaky, 517.

Alkenes: Behaviour of certain substituted — towards the Meinel colour test. F. B. LaForge and F. Acree, Jr., 614.

All-India Institute of Hygiene and Public Health: Report for 1938. J. B. Grant, 225.

- *Alloy(s):** lead —; Simple test for arsenic in. R. G. Robinson, 159.
- Lead-sodium** — as a drying agent. H. Soroos, 191.
- *nickel-**; Determination of aluminium, magnesium or beryllium in —. R. C. Chirnside, L. A. Dauncey and P. M. C. Proffitt, 446.
- rich in tin; Determination of tellurium in — by volatilisation. W. T. Pell-Walpole, 434.
- Almonds:** oil of bitter —; Variations in. 503.
- *Alum Cake:** free acid in —; New method of determining. J. R. Simmons, 456.
- *Alumina** for estimating carotene. W. M. Seaber, 645.
- in silicates; Direct determination of —. E. W. Koenig, 130.
- iron in —; Colorimetric determination of. L. Roelen, 187.
- Aluminium:** Aluminium oxide in metallic — determined by means of cupric ammonium chloride. T. Nakamura and S. Yamazaki, 68.
- Colorimetric estimation of silica and its use in metallurgy of —. P. Urech, 131.
- Determination of — as phosphate. 339.
- *in nickel alloys;** Determination of —. R. C. Chirnside, L. A. Dauncey and P. M. C. Proffitt, 446.
- in presence of iron and phosphoric acid; Determination of —. G. Balanescu and M. D. Motzoc, 188.
- iron in —; Colorimetric determination of. L. Roelen, 187.
- Separation of cadmium from zinc by precipitation with —. F. E. Townsend and G. N. Cade, 310.
- Spot tests for —. E. A. Kocsis, 190.
- *Aluminium Hydrous Oxide:** Precipitation of — and its solubility in ammonia. E. B. R. Prideaux and J. R. Henness, 83.
- Aluminium Oxide** in metallic aluminium determined by means of cupric ammonium chloride. T. Nakamura and S. Yamazaki, 68.
- Amines:** 3,5-Dinitro-4-methylbenzazide as reagent for identifying —. P. P. T. Sah, 125.
- Identification of — with *p*-nitrobenzazide and *p*-nitrophenyl isocyanate as reagents. P. P. T. Sah, 374.
- Amino Acids** and their compounds; Microscopy of —. IV. Picrolonates. R. Dunn, K. Inouye and P. L. Kirk, 252.
- content of human serum proteins; Basic —. Influence of ingestion of arginine on composition of serum proteins. R. J. Block, 303.
- Experiments on —. R. L. M. Synge, 238.
- Naphthalene- β -sulphonic acid as reagent for —. M. Bergmann and W. H. Stein, 237.
- Semi-micro-estimation of —. H. R. Ing and M. Bergmann, 57.
- Separation of — by means of their N-acetyl derivatives. R. L. M. Synge, 238.
- 4-Aminodiphenylamine:** Separation and determination of —. I. S. Shupe, 308.
- 2-Aminoethanol:** Identification of —. B. Keiser, 474.
- 4-Amino-2-Methyl-1-Naphthol:** Vitamin K activity of —. A. D. Emmett, O. Kamm and E. A. Sharp, 305.
- Amino Nitrogen:** Behaviour of some uramido acids in the nitrous acid method for determining —. A. G. Gornall and A. Hunter, 467.
- in malt extracts; Determination of —. S. R. Snider, 464.
- o*-Aminophenol** in *p*-aminophenol; Estimation of — by fluorescence analysis. W. Seaman, A. R. Norton and O. E. Sundberg, 581.
- Separation of *p*-aminophenol, *p*-methylaminophenol and —. I. S. Shupe, 667.
- p*-Aminophenol:** *o*-Aminophenol in — estimated by fluorescence analysis. W. Seaman, A. R. Norton and O. E. Sundberg, 581.
- Separation of *p*-methylaminophenol, *o*-aminophenol and —. I. S. Shupe, 667.
- Ammonia:** Detection of — with organic nitro compounds. R. Fischer, 135.
- in toilet preparations. 165.
- Mechanism of microbial oxidation of —. G. Gopala Rao and W. V. Sundara Rao, 524.
- Proceedings from sale of —. 503.
- *Solubility** of aluminium hydrous oxide in —. E. B. R. Prideaux and J. R. Henness, 83.
- Anaesthetic** chloroform; Chromic oxidation of —. 124.
- ether. B.P. Sub-Committee recommendation. 167.
- Aneurin** estimation by a modification of the Melnick-Field method. A. D. Emmett, G. Peacock and R. A. Brown, 661.
- in blood; Estimation of —. II. Further modification of Meiklejohn's method. H. M. Sinclair, 182.
- in foods; Distribution of — determined by chemical analysis. M. Pyke, 180.
- in urine; Chemical estimation, stability and form of —. D. Melnick and H. Field, Jr., 56.
- Thiochrome method for estimating —, with survey of — content of wheats. R. G. Booth, 610.
- *Anhydrite** as desiccating agent. H. Hawley, 644.
- Aniline** vapour in industry; Methods for detecting —. D.S.I.R. Leaflet No. 11. 162.
- Animal** experiments; Vitamin-free diets for —. A. L. Bacharach, 573.
- organs; Cobalt in —. 34.
- Animals:** Minimum lethal dose of selenium as sodium selenite for certain —. W. T. Miller and K. T. Williams, 613.
- Anions:** Salts of complex cations applied to microscopical detection of —. Mono-aquopentamminocobaltic chloride. W. A. Hynes and L. K. Yanowski, 70; 1,2-Chloro-aquotetramminocobaltic chloride. L. V. Yanowski and W. A. Hynes, 190.
- Anti-Grey Hair Vitamin** for silver foxes; Studies on —. G. Lunde and H. Kringstad, 366.

- Antimony**: Gravimetric determination of — by means of 8-hydroxyquinoline. T. I. Pirtea, 188.
- *in organic compounds; Micro-tests for —. C. L. Wilson, 405.
- *in tin-antimony oxides. A. G. Dunbar-Poole, 453.
- Separation of zinc from —. 381.
- Antimony Trichloride**: Behaviour of olive oil and other oils with —. W. H. Dickhart, 421.
- Antineuritic** value of parboiled rice. I. A. Simpson, 461.
- Antiscorbutic** values of fruits and vegetables. M. Olliver, 662.
- Aphis**: cabbage —; Rate of production. 105.
- Apparatus**: Abstracts: 73, 136, 191, 253, 382, 435, 534, 581, 670.
- Apple** leaves; Copper in —. 292.
- Apples**: Arsenic in —. 229.
- *Composition of —. 550, 554.
- Applied Chemistry**: Annual Reports of Progress of —. Vol. XXIV for 1939. (Review), 436.
- Thorpe's Dictionary of —. 4th Ed. (Review), Sir J. F. Thorpe and M. A. Whiteley, 583.
- Appointments**: Official —. 27, 161, 411, 456, 566.
- ***Apricots** and apricot pulp; Composition of —. 552, 554.
- Arachidonic Acid**: Isolation of —. G. Y. Shinowara and J. B. Brown, 576.
- Arachis Oil**: Thermo-stability of —. 301.
- Arc Light**: Poisonous fumes from —. 292.
- Argemone Mexicana Oil**: Epidemic dropsy and —. 226.
- Argentometric** determination of cyanide with diphenylcarbazone as indicator. R. Ripan-Tilici, 315.
- Arginine**: Estimation of — by means of flavianic acid. H. B. Vickery, 236.
- Influence of ingestion of — on composition of serum proteins. R. J. Block, 303.
- Aristoquin**: Application of herapathite reaction to —. M. Wagenaar, 111.
- Armant**: Temples of —. (Review) Sir R. Mond and O. H. Myers, 672.
- Aromatic halogenonitro** compounds; Identification of — as piperidyl derivatives. M. K. Seikel, 666.
- hydrocarbons in air; Detection of —. G. R. Gilbert and R. E. Tannich, 666.
- Arrack**: Examination of — in Ceylon. 652.
- Arrow Poisons**: African —. *Adenium somalense* Balf. fil. M. Hartmann and E. Schlitter, 428.
- Arsenic** compounds; Precipitation reactions of organic —. M. Péronnet and R. H. Rémy, 247.
- content of fresh vegetables; Limit for —. 291.
- in biological material; Rapid method for accurate estimation of minute quantities of —. J. Bodnár, O. Szep and V. Cieleszky, 379.
- in coal and coke; Determination of —. 649.
- in cremation remains. 601.
- Arsenic—continued**
- *in lead alloys; Simple test for —. R. G. Robinson, 159.
- in organic and inorganic compounds; Determination of —. D. T. Lewis and V. E. Davis, 531.
- *in organic compounds; Micro-tests for —. C. L. Wilson, 405.
- in pears and apples. 229.
- in soils and waters. 34.
- in soils and waters in the Waiotapu Valley, New Zealand. R. E. R. Grimmer and I. G. McIntosh, 613.
- in urine; Colorimetric semi-micro-determination of —. J. V. Harspe, 54.
- Industrial poisoning with —. 291.
- nephelometric determination of — with Bougault's reagent; Permanent artificial standards for. J. Thuret, 129.
- Quantitative test for —. B. P. Subcommittee recommendations. 167.
- Separation of zinc from —. 381.
- Arsenical** insecticides and wine. 166.
- Arsine**: Detection of — in industry. D.S.I.R. Leaflet No. 9, 354.
- Arylation** of oils and fats. Synthesis of tolyl stearic acid and its esters. W. Kimura and J. Tsurugi, 186.
- Ascorbic Acid** as substrate in oxidase measurements. B. D. Egell and F. Gerhardt, 469.
- content of rose hips. W. Goldberg and E. O'F. Walsh, 304.
- content of tablets; Determination of —. E. C. M. J. Hollman, 365.
- in animal tissue; Combined —. P. Holtz, 369.
- in blood serum; Factors preventing oxidation of —. E. M. Mystkowski and D. Lasocka, 59.
- in plant foodstuffs; Combined —. J. C. Pal, B. C. Guha, and P. N. Sen-Gupta, 522, 523.
- oxidase from summer crook-neck squash (*C. pepo condensata*). P. L. Lovett-Janison and J. M. Nelson, 572.
- solutions; Oxalate formation in —. A. E. Jurist and W. G. Christiansen, 184.
- Ascorbigen**: Concentration of — from cabbage. B. Ghosh and B. C. Guha, 523.
- Ashing** soft tissues preliminary to determining cations. M. V. Buell, 53.
- Asparagus**: Enzyme activity in frozen —. M. A. Joslyn and C. L. Bedford, 468.
- Asphalt**: Artificial weathering of —. 598.
- Atmospheric** corrodibility of ferrous materials; Accelerated spray test for determining —. T. Swinden and W. W. Stevenson, 670.
- pollution; Conference on investigation of —. 462.
- Atomic Weights**: Tenth Report of Committee on —. 415.
- Atoms**: Fifth Report of Committee on —. 568.
- Atractoscion acquideus C. and V.**: vitamin A content of visceral organs of —; Seasonal variations in. C. J. Molteno and W. S. Rapson, 59.
- Avitaminosis A** in man; Experimental —. K. H. Wagner, 426.

B

- B. coli** in crab and lobster meat. 227; in vegetables, 227.
- Bacon** factory; Refuse from —. 462.
Function of nitrate, nitrite and bacteria in the curing of —. 355.
- Bacteria**: Classification and identification of —, especially of beer types of —. J. L. Shimwell, 612.
from chlorinated waters. M. Levine, P. Carpenter and J. M. Coblentz, 370.
Function of — in curing of bacon and hams 355.
Stability of — in relation of pH. J. G. Baumgartner and G. G. Knock, 371.
- Bacterial** content of paper-board; Routine determination of —. L. C. Cartwright and S. S. Epstein, 612.
- Bacteriological** analysis. Abstracts: 62, 120, 243, 370, 428, 470, 524, 574, 612.
- Bacteriology** of fresh water. 102.
Textbook of —. 3rd Ed. (Review). R. W. Fairbrother, 673.
- Baking Powder** and self-raising flour. 650.
Labelling of —. 291.
- Bambara Groundnut**: J. M. Holm and B. W. Marloth, 608.
- Bamboo** leaf wax. M. Tsujimoto, 187.
shoots; Poisonous substance in —. 37.
- Banana** starch; Constitution of —. E. G. E. Hawkins, J. K. N. Jones and G. T. Young, 464.
- Bangalore**: Report of Public Analyst for Municipality of — for 1939-40. G. N. Murty, 353.
- Barbiturates** in blood; Estimation of —. G. A. Levvy, 471.
- Barium**: Detection of — by spot reactions. F. Feigl and W. Aufricht, 251.
Modified iodate method for determining —. F. C. Guthrie, 434.
Volumetric determination of small quantities of — with barium rhodizonate as indicator. C. C. Miller, 477.
- *Barium Carbonate**: carbon dioxide in —; Determination of. 641.
- Barium Rhodizonate** as indicator in the volumetric determination of small quantities of barium and sulphate. C. C. Miller, 477.
- Barium Sulphate**: washing and isolation of — in the micro-determination of sulphur; New method for. R. Grangaud, 316.
- Barley** plant; Sucrase activity in —. H. K. Archbold, 520.
vitamin B₁ content of —; Effect of soil treatment on. P. C. Leong, 59.
- Basins**: B.S.I. standard dimensions for silica —. 41.
- Batley**: Appointment of F. W. M. Jaffé as Public Analyst for Borough of —. 411.
- Battersea**: Appointment of A. H. M. Muter as Deputy Public Analyst for Metropolitan Borough of —. 161.
- Bauhinia variegata** seeds; Fatty oil from —. S. V. Puntambeka and S. Krishna, 421.
- Beam's Test** for hashish; Application of —. 166.
- Beckenham**: Appointment of J. W. Flint as Deputy Borough Analyst for —. 161.
- Beddington and Wallington**: Appointment of H. A. Williams as Public Analyst for Borough of —. 411.
- Beef**: corned —; Hydrogen sulphide in. A. Elias, 605.
*fat in lard; Detection of —. K. A. Williams, 596. Erratum, 645.
fat; Thermo-stability of —. 301.
- Beer**: Lead in —. 28.
Oxidation turbidities in — and their causes. E. Singruen, 569.
*Succinic acid in —. R. L. Andrew and L. G. Neubauer, 455.
Sulphur dioxide in —. 599.
types of bacteria; Classification and identification of —. J. L. Shimwell, 612.
- Bellier Test** and the Blarez test. Application to olive and other oils. R. Marcille, 170.
- *Benzedrine**: Detection and estimation of —. E. T. Illing, 3.
- Benzidine**: Reaction of copper with —. G. Spacu and C. G. Macarovici, 248.
- *Benzoic Acid**: Determination of —. R. W. Sutton and O. Hitchen, 502.
Little-known reaction for —. N. Schoorl, 374.
- Benzyl Sulphanilamide**: Determination of —. 212.
- Berberis vulgaris** and *agrifolium*: alkaloids in preparations of —; Determination of. K. Brunner and H. Neugebauer, 47.
- *Beryllium** in nickel alloys; Determination of —. R. C. Chirnside, L. A. Dauncey and P. M. C. Proffitt, 446.
- Bexley**: Appointment of F. W. F. Arnaud as Public Analyst for Borough of —. 27.
- Bicycles**: Deciphering numbers of —. 652.
- *Bilberries**: Composition of —. 551, 554.
- Biochemical** analysis: Abstracts: 51, 112, 176, 235, 302, 366, 425, 466, 519, 570, 609, 659.
- Biochemistry**: Introduction to —. 2nd Ed. (Review), W. R. Fearon, 387.
of micro-organisms. Emodic acid and ω -hydroxyemodin, metabolic products of *Penicillium cyclopium* Westling. W. K. Anslow, J. Breen and H. Raistrick, 470.
- Biological** fluids; Colorimetric estimation of quinine in —. R. O. Prudhomme, 239.
fluids; Determination of sodium in —. M. C. Darnell, Jr., and B. S. Walker, 478.
fluids; Estimation of small amounts of sulphur as sulphates in —. A. D. Marenzi and R. F. Banfi, 366.
materials; Bibliography on metals in foods and —. I. Cobalt, 513. II. Nickel, 603.
materials; Determination of magnesium in —. J. P. Nielsen, 176.
materials; Determination of small quantities of molybdenum in —. D. Bertrand, 177.
materials; Determination of tungsten in —. J. C. Aull and F. W. Kinard, 660.
materials; Estimation of nicotinic acid in — by means of photoelectric colorimetry. D. Melnick and H. Field, 571.

Biological—continued

- materials; Rapid method for accurate estimation of minute quantities of arsenic in —. J. Bodnár, O. Szep and V. Cielezsky, 379.
- substances; Photometric estimation of silicic acid in —. J. Bodnár and T. Török, 240.
- Biotin**: Quantitative test for —. E. E. Snell, R. E. Eakin and R. J. Williams, 524.
- Birds**: Vitamin A and carotenoids in liver of —. H. B. Jensen and T. K. With, 179.
- Birmingham**: Report of City Analyst (H. H. Bagnall) for the year 1939, 597; for Second quarter, 1940, 565.
- Bismuth** determined by precipitation as quinaldine iodobismuthite. J. R. Hayes and G. C. Chandless, 129.
- in quinine iodobismuthate and its injectable preparations; Determination of —. G. N. Thomis and G. P. Koponaris, 49.
- New method for volumetric determination of —. L. Malaprada, 249.
- Separation of zinc from —. 381.
- Bitter Almond Oil**: Variations in —. 503.
- Bituminous** road mixtures; B.S.I. Methods for sampling and examining —. 654.
- Biuret Reaction** in estimating serum proteins. H. W. Robinson and C. G. Hogden, 466.
- ***Blackberries**: Composition of —. 551, 554.
- ***Blackcurrants**: Composition of —. 548, 553.
- Blarez Test** and Bellier test. Application to olive and other oils. R. Marcille, 170.
- Blast Furnace** slag; Utilisation of —. 598.
- Bleach** ointment. G. H. Macmorran, 423.
- ointment. B.S.I. specification for —. 463.
- ointment. (Legal Notes), 30.
- *ointments. D. D. Moir, 154.
- Bleaching** of flour with chlorine; Detection of —. D. B. Scott, 605.
- Bleaching Powder**: False trade description of —. (Legal Note), 565.
- *Reaction between 2,2'-dichlorodiethyl sulphide (mustard gas) and —. A. G. Lipscomb, 100.
- test for aniline vapour. 162.
- Blood**: acetone bodies and small amounts of mercury in —; Method for estimating. L. A. Crandall, 467.
- aneurin in —; Estimation of. II. Further modification in Meiklejohn's method. H. M. Sinclair, 182.
- barbiturates in —; Estimation of. G. A. Levy, 471.
- carbon monoxide in —; Estimation of. C. Scholten, 471.
- distillates; Determination of alcohol in presence of acetone in —. 600.
- iodine in —; Permanganate acid ashing micro-method for determining. D. S. Riggs and E. B. Man, 579.
- lead in —; Influence of calcium, phosphorus and vitamin D on. A. E. Sobel and Others, 235.
- Natural fat glycerol in — determined with periodate. L. Voris, G. Ellis and L. A. Maynard, 425.
- nicotinic acid in —; Estimation of. E. Kodicek, 522.

Blood—continued

- pyruvic acid in —; Stabilisation and determination of. E. Bueding and H. Wortis, 426.
- serum; Factors preventing oxidation of ascorbic acid in —. E. M. Mystkowski and D. Lasocka, 59.
- stains; "Grouping" of —. 460.
- stains; Identification of —. 652.
- sugar levels in rats receiving the cataractogenic sugars galactose and xylose. W. J. Darby and P. L. Day, 520.
- sulphur in —; Estimation as sulphates. 366.
- Boiler Water** treatment; Application of hydrogen ion concentration to —. G. W. Bond, 429.
- Bone**: Lead in —. 165.
- lead in —; Influence of calcium, phosphorus and vitamin D on. A. E. Sobel and Others, 235.
- Book Reviews**: Anderson, A. K. Essentials of Physiological Chemistry. 76.
- Appleyard, F. N., and Lyons, C. G. Practical Pharmaceutical Chemistry. 4th Ed. 436.
- Bernal, J. D. Social Function of Science. 140.
- British Encyclopaedia of Medical Practice. (1) Toxicology. (2) Urine Examination. (3) Vitamins. Vol. 12. 139.
- Brode, W. R. Chemical Spectroscopy. 137.
- Burton, D., and Robertshaw, G. F. Sulphated Oils and Allied Products—Their Chemistry and Analysis. 318.
- Clark, W. Photography by Infra-Red: Its Principles and Applications. 480.
- Cochrane, J. A. Lavoisier. 320.
- Conway, E. J. Micro-Diffusion Analysis and Volumetric Error. 254.
- Cumming, A. C., and Kay, S. A. A Text-book of Quantitative Chemical Analysis. 7th Ed. 138.
- Dorsey, N. E. Properties of Ordinary Water Substance. 621.
- Egloff, G. Physical Constants of Hydrocarbons. Vol. II. 585.
- Engelder, C. Calculations of Quantitative Analysis. 674.
- Ephraim, F. Inorganic Chemistry. 3rd Eng. Ed. by P. C. L. Thorne and A. M. Ward. 584.
- Evans, R. C. An Introduction to Crystal Chemistry. 75.
- Fairbrother, R. W. Text Book of Bacteriology. 3rd Ed. 673.
- Fearon, W. R. An Introduction to Biochemistry. 2nd Ed. 387.
- Fiene, F., and Blumenthal, S. Handbook of Food Manufacture. 255.
- Findlay, A. Chemistry in the Service of Man. 5th Ed. 535.
- Gordon, N., and Trout, W. E., Jr. Introductory College Chemistry, 2nd Ed. 537.
- Hibben, J. H. The Raman Effect and its Chemical Applications. 192.
- Hilditch, T. P. The Chemical Constitution of Natural Fats. 319.
- Hind, H. Lloyd. Brewing Science and Practice. Vol. II. 482.
- Holmstrom, J. E. Records and Research in Engineering and Industrial Science. 322.

Book Reviews—continued

- Mann, F. G., and Saunders, B. C. Introduction to Practical Organic Chemistry. 74.
- Mason, R. D. Fat, Total Solids and Moisture. 75.
- Matagrín, A. Manuel du Savonnier. 389.
- Mineral Water Trade Year Book. 142.
- Mond, Sir R., and Myers, O. H. Temples of Armant. 672.
- Moore, F. J. A History of Chemistry. 3rd Ed. 386.
- Parkes, G. D., and Mellor, J. W. Mellor's Modern Inorganic Chemistry. 9th Ed. 479.
- Plimmer, V. G. Food Values at a Glance. 388.
- Radley, J. A. Starch and its Derivatives. 481.
- Radley, J. A., and Grant, J. Fluorescence Analysis in Ultra-Violet Light. 3rd Ed. 194.
- Reilly, J., and Rae, W. N. Physico-Chemical Methods. Vol. I. Measurement and Manipulations. Vol. II. Practical Measurements. 384.
- Rhodes, H. T. F. Forensic Chemistry. 621.
- Robinson, R. A., and Ives, R. Bell's Sale of Food and Drugs. 10th Ed. 385.
- Sachanen, A. N. Conversion of Petroleum. 671.
- Sand, H. J. S. Electrochemistry and Electrochemical Analysis. Vol. II. 620.
- Society of Chemical Industry. Annual Reports of Progress of Applied Chemistry. Vol. XXIV for 1939. 436.
- Society of Chemical Industry Food Group. Vitamin E. 77.
- Strong, R. K. Kingzett's Chemical Encyclopaedia, 6th Ed., 537.
- Thompson, M. de K. Theoretical and Applied Electrochemistry, 3rd Ed. 193.
- Thorpe, Sir J. F., and Whiteley, M. A. Thorpe's Dictionary of Applied Chemistry. 4th Ed. 583.
- Tweeney, C. F., and Hughes, L. E. C. Chambers's Technical Dictionary. 622.
- Vogel, A. I. Text-book of Quantitative Inorganic Analysis. 195.
- Walter, F. G. The Manufacture of Compressed Yeast. 586.
- Weeks, M. E. Discovery of the Elements. 4th Ed. 321.
- Weinig, A. J., and Schoder, W. P. Technical Methods of Ore Analysis for Chemists and Colleges. 11th Ed. 194.
- Weiser, H. B. Colloid Chemistry—A Text-book. 536.
- Yoe, J. H. A Laboratory Manual of Qualitative Analysis. 254.
- Boric Acid** colour reaction of flavone derivatives. C. W. Wilson, 66.
- Boron** in soils and plants determined by means of quinalizarin. K. C. Berger and E. Truog, 123.
- Bougault's Reagent**: nephelometric determination of arsenic with —; Permanent artificial standards for. J. Thuret, 129.
- Bran**: Rice — as a raw material of oil. T. Hidaka, 44.
- Brass** electroplating baths; Determination of free cyanide in —. W. M. McNabb and S. Heiman, 315.
- Brassicasterol**: the characteristic sterol of rapeseed oil. E. Fernholz and H. E. Stavely, 572.
- Bread** and its ingredients; Determination of iron in —. C. Hoffman, T. R. Schweitzer and G. Dalby, 569.
- Aromatic principle in —. II. Isolation of aromatic principle in rye bread. E. Komm and G. Lehmann, 360; III. Colorimetric estimation of diacetyl, acetoin and butylene glycol. E. Komm and J. Flügel, 361.
- brown —; Standard fibre content of. 649.
- standardisation. 599, 649.
- wholemeal —; Standard fibre content of. 649.
- Brewing** industry; Thermal death time of micro-organisms encountered in —. S. S. Epstein and F. D. Snell, 524.
- Science and Practice. Vol. II. (Review), H. Lloyd Hind, 482.
- Brewing Effluents Committee**: Report of —. 356.
- Bricks**: Sand-lime —. 598.
- Brierley Hill**: Appointment of E. V. Jones as Public Analyst and F. Dixon as Deputy Analyst for Urban District of —. 411.
- Brisling**: Significance of bacterial count in determining freshness of — and raw materials of canned —. E. Aschehoug and R. Vesterhus, 415.
- Bromide**: Detection of —. D. Hart and R. Meyrowitz, 532.
- Bromide**: iodide in mineral waters containing —; Determination of. E. Müller and W. Stumpf, 245.
- Bromides** in presence of chlorides; Determination of —. A. Denoël, 314.
- British Leather Manufacturers' Research Association**: Decontamination of leather contaminated with mustard gas. 39.
- British Non-Ferrous Metals Research Association**: Quantitative spectrographic analysis with the microphotometer, 295.
- British Pharmacopoeia**, 1932: Alterations and Amendments, 457.
- Second Addendum to, 457.
- British Pharmacopoeia Commission**: Report of Committee on General Chemistry. No. 14. 167.
- Report of Committee on Pharmacy and Pharmacognosy. No. 13. 107.
- British Standards Institution Specifications**: A.R.P. No. 27. Testing incombustible material resistant to incendiary bombs. 297.
- A.R.P. No. 40. Bleach ointment (anti-gas ointment No. 1), 463.
- No. 593. General purpose laboratory thermometers. 463.
- No. 598. Methods for sampling and examination of bituminous road mixtures. 654.
- No. 611. Petri dishes. 463.
- No. 875. Dimensions for silica basins, crucibles and capsules. 41.
- No. 894. Determination of flow and drop points of fats and allied substances. 418.
- No. 895. Methods for microbiological examination of butter. 359.
- No. 914. Tests for laboratory porcelain. 516.
- War emergency standards, 463.

- Bromate**: Volumetric determination of —. C. Mahr and H. Ohle, 189.
- Bromine** absorption as means of determining unsaturation in aliphatic hydrocarbon mixtures. J. B. Lewis and R. B. Bradstreet, 576. in foodstuffs; Micro-determination of —. W. P. Ford and Others, 617.
- Bromley**: Appointment of F. W. F. Arnaud as Public Analyst for Borough of —. 27. Appointment of J. W. Flint as Deputy Borough Analyst for —. 161.
- Bromothymol Blue** test for mastitis. 35.
- Buffalo milk fat**; Turkish —. A. Heiduschka and F. Cicekdagi, 297.
- Buffer** solution; Carbonate veronal — covering pH 7.5 to 10.7. E. J. King and G. E. Delory, 469.
- *Universal — mixture. H. T. S. Britton, 220.
- Building Research Board**: Report for 1939, 598.
- Buildings**: Cleaning of —. 598.
- Burton-upon-Trent**: Appointment of R. Mallinder as Public Analyst and Agricultural Analyst for Borough of —. 411.
- Butter**: B.S.I. standard for methods of microbiological examination of —. 359.
- *carotene in —; Determination of. 276. iron and copper in —; Determination of. G. M. Moir and E. D. Andrews, 657. yellow; Effect of oral administration of — on growth of rats. J. White, 467.
- n-Butyl-p-Hydroxybenzoate** as preservative. 291.
- Butylene Glycol**: Colorimetric estimation of —. E. Komm and J. Flügel, 361.
- C**
- Cabbage** aphids; Rate of production. 105. Concentration of ascorbigen from —. B. Ghosh and B. C. Guha, 523.
- ***Cadmium**: copper in small quantities in presence of —; Electrolytic determination of. J. G. Fife, 562.
- *plating solutions; Analysis of —. G. S. Smith, 87.
- *Quinaldine acid as reagent for separation of copper and —. A. J. Lindsey and R. J. Shennan, 636. separated from zinc by precipitation with aluminium. F. E. Townsend and G. N. Cade, 310. Test for —. E. Eegriwe, 248.
- Calcium**: Detection of — with organic nitro compounds. R. Fischer, 135. Influence of — on lead in blood and bone. A. E. Sobel and Others, 235. manganese in organic material containing large amounts of —; Estimation of. T. W. Ray, 570. metabolism; Studies in —. 225.
- Calcium Hydroxide**: Precipitation of natural tannins with —. V. Nemec, 246.
- Calcium Hypochlorites**: Stability of —. A. Guillaume and Y. Nicolas, 130.
- Camphorated Oil**: Adulterants in —. 600.
- Canadian wheat**; Selenium in —. J. G. Malloch, 430.
- Canarium Commune L.** seeds; Oil from —. A. Steger and J. van Loon, 230.
- Candles**: lac in glazed —; Volumetric estimation of. N. M. Molnar and J. Grumer, 175.
- Cannabidiol**: Isolation from Egyptian hashish. A. Jacob and A. R. Todd, 466.
- Cannabinol**: Observations on structure of —. A. Jacob and A. R. Todd, 466.
- Cannabis Indica**: A. Jacob and A. R. Todd, 466.
- Canned** brisling; Significance of bacterial count in determining freshness of raw materials of —. E. Aschehoug and F. Vesterhus, 417. fish; Examination of oil from —. 171. fish; Lead, tin and zinc in —. 28. food; Lead in —. 459. fruit and vegetables; Drained weight of English —. 511. fruit; Behaviour of traces of sulphur in —. 512. fruit; Hydrogen "swells" in —. T. P. Hoar, 43; Study of —. 512. vegetables; Hydrogen ion concentration of —. 512. vegetables; Zinc in —. 651.
- Canning** industry; Olive specifications for the Norwegian —. E. Mathiesen, 296. of meat in the home by the pressure cooker; Effectiveness of heat penetration in —. C. I. Nelson and D. Berrigan, 120.
- Cans**: Heat penetration in rotating —. 512.
- Cape Salmon**: vitamin A content of visceral organs of —; Seasonal variations in. C. J. Molteno and W. S. Rapson, 59.
- Capsules**: B.S.I. standard dimensions for silica —. 41.
- Carbohydrate** characterisation. I. Oxidation of aldoses by hypiodite in methanol. II. Identification of seven aldo-monosaccharides as benzimidazole derivatives. S. Moore and K. P. Link, 419.
- Carbohydrates**: Metabolism of potato —. 224.
- Carbolic Acid**: Is it a drug? (Legal Notes), 221.
- Carbolic Ointment**: (Legal Notes), 222.
- Carbon**: Isotopes of —. 568. metabolism of the Kelsey plum; Role of sorbitol in —. I. Donen, 178. Wet combustion method for determining — and oxygen consumed. B. E. Christiansen and J. F. Facer, 64.
- ***Carbon Dioxide** in carbonates; Micro-titrimetric determination of —. A. F. Colson, 638.
- Carbon Disulphide** in air; Determination of —. F. F. Moorhead, 525.
- Carbon Monoxide**: Elimination of traces of — from hydrogen. K. Kawakita and B. Ichiiyanagi, 575. in blood; Estimation of —. C. Scholten, 471. in industry; Detection of —. D.S.I.R. Leaflet No. 7, 223. Separation of para-hydrogen from —. E. Bergmann, L. Farkas and L. Sandler, 244.
- Carbonate Veronal Buffer Solution** covering pH 7.5 to 10.7. E. J. King and G. E. Delory, 469.
- ***Carbonates**: carbon dioxide in —; Micro-titrimetric determination of. A. F. Colson, 638.

- Carboxide** as fumigant for foodstuffs. 599.
- Cardiff**: Report of the City Analyst for 1938. S. Dixon, 28.
- Carotene**: Adsorption method for determining pure. G. S. Fraps, A. R. Kemmerer and S. M. Greenberg, 609.
- ***Alumina** for estimating ——. W. M. Seaber, 645.
- *and allied pigments; Commercial determination of —, with special reference to dried grass and other leafy materials. W. M. Seaber, 266.
- *and allied pigments; Constitution and physiological significance of —. R. A. Morton, 263.
- *and allied substances in foods and feeding stuffs. 263.
- content of yolk of hens' eggs. B. Sjollem and W. F. Donath, 521.
- Coupled oxidation of fat and — by — oxidase. J. B. and R. J. Sumner, 571.
- Estimation of — with the photoelectric colorimeter. C. J. Koehn and W. C. Sherman, 303.
- in presence of lycopene; Determination of —. G. S. Fraps, A. R. Kemmerer and S. M. Greenberg, 608.
- Carotenoids** in the liver of mammals, birds, reptiles and man. H. B. Jensen and T. K. With, 179.
- Carrots**: Comparison of analyses of red and white —. 462.
- Carshalton**: Appointment of E. Hinks as Public Analyst and D. D. Moir as Deputy Analyst for Urban District of —. 411.
- Cartridges**: Size of shot in —. 459.
- Carviolin**: Constitution of —. H. G. Hind, 470.
- Casein**: Vitamin A content of "light white" —. M. K. Maitra and T. Moore, 116.
- wool; Determination of addition of — to true woollen cloths. J. Straub and G. J. Van Amerongen, 582.
- Cassia**: Quantitative determination of powdered —. A. H. Saber, 421.
- Cast Iron**: molybdenum in —; Determination of. C. Sterling and W. P. Spuhr, 311.
- Castleford**: Appointment of F. W. M. Jaffé as Public Analyst for Urban District of —. 411.
- Catechin** and catechin-tans in gambir; Determination of —. C. J. van Hulssen and D. R. Koolhaas, 127.
- Catenarin**: Molecular constitution of —. W. K. Anslow and H. Raistrick, 574.
- Cations**: Ashing of soft tissues preliminary to determination of —. M. V. Buell, 53.
- Salts of complex — applied to the microscopical detection of anions. Mono-aquopentamminocobaltic chloride. W. A. Hynes and L. K. Yanowski, 70; 1-2-Chloro-aquotetramminocobaltic chloride. L. V. Yanowski and W. A. Hynes, 190.
- Cattle**: Ragwort feeding to —. 35.
- Caustic Soda** poisoning. 459.
- Ceilings**: Pattern-staining of —. 598.
- Cell** for electroanalysis, especially of soils. A. W. Marsden, 435.
- Cellulose**: Micro-chemical determination of reducing power of — (micro-copper, micro-ferricyanide and micro-iodine numbers). E. Geiger and G. Müller, 618.
- spray paints; Lead compounds in —. 292.
- Cement**: Fineness of Portland —. 598.
- Centaurea scabiosa***: Study of —. C. Charaux and J. Rabaté, 302.
- Centrifuge**: Efficiency of Wells air —, as determined by air-washing technique. K. MacDonald, 470.
- Centrifuging**: Separation of the components of a mixture by fractional —. A. Gourevitch, 136.
- Cerate Oxidimetry**: Determination of iodine in thyroid and its preparations by —. W. W. Hilty and D. T. Wilson, 174.
- Stability of solutions. G. F. Smith and C. A. Getz, 532.
- Cerberum odallum*** kernels; Constants of oil from —. 37.
- Cereals**: Phytic acid and the rickets-producing action of —. D. C. Harrison and E. Mellanby, 113.
- Cerebro-Spinal Fluid**: vitamin B₁₂ in —; Estimation of. H. M. Sinclair, 181.
- Cerium** determination by means of 8-hydroxyquinoline. R. Berg and E. Becker, 312.
- in cerous salts; Estimation of —. J. Plank, 434.
- Cerous Salts**: cerium in —; Estimation of. J. Plank, 434.
- Ceylon**: Report of Government Analyst for 1939. J. V. Collins, 652.
- Chambers's** Technical Dictionary. (Review), C. F. Tweeney and L. E. C. Hughes, 622.
- Chaulmoogra Oils**: Analysis of —. *Hydnocarpus wightiana* oil. H. I. Cole and H. T. Cardoso, 45; *Hydnocarpus anthelmintica* oil and *Taraktogenos Kurzii* oil. 231.
- Cheese**: Cream —. Ministry of Agriculture and Fisheries Advisory Leaflet No. 222, 456.
- Milk —. 565.
- "spreads", 597.
- Standards for — in New South Wales. 650.
- Vitamin A content of —. A. W. Davies and T. Moore, 58.
- Chemical Encyclopaedia**; Kingzett's —. 6th Ed. (Review), Revised by R. K. Strong, 537.
- Chemicals**: B. P. Sub-Committee on General Organic — and on Inorganic —. Recommendations. 167.
- Chemistry**: History of —. 3rd Ed. (Review), F. J. Moore, 386.
- in the Service of Man. 5th Ed. (Review), A. Findlay, 535.
- Introductory College —. 2nd Ed. (Review), N. Gordon and W. E. Trout, Jr. 537.
- See also **International Union of Chemistry**.
- Chinese** coconut oil; Composition of fatty acids in —. H. Nobori, 658.
- Chislehurst and Sidcup**: Appointment of F. W. F. Arnaud as Public Analyst for Urban District of —. 411.
- Chloralose** in urine; Detection of —. M. P. Cheramy, 471.
- Chlorate**: Volumetric determination of —. C. Mahr and H. Ohle, 189.

- Chloride:** Detection of —. D. Hart and R. Meyrowitz, 532.
iodide in mineral waters containing large quantities of —; Determination of. E. Müller and W. Stumpf, 245.
- Chlorides:** bromides in presence of —; Determination of. A. Denoël, 314.
in presence of thiocyanates and cyanides, Determination of —. A. Slooff and D. van Duyn, 250.
in tomato products; Rapid method for determining —. L. M. Beacham, 109.
manganese in organic material containing large amounts of —; Estimation of. T. W. Ray, 570.
- Chlorine** in coal and coke; Determination of —. 649.
in effluents; Colorimetric determination of free —. F. J. Hallinan, 578.
in fat of flour; Determination of —. D. B. Scott, 605.
- Chloroform:** Chromic oxidation of anaesthetic —. 124.
- Chlorophyllase:** C. A. Weast and G. Mackinney, 426.
- Chocolat:** diabetic —; *d*-Sorbitol in. 651.
*Preparation of a standard microscope slide with special reference to —. H. C. Lockwood, 97.
- Chondrodendron Species:** Alkaloids of —. H. King, 518.
- *Chromates** in sewage liquors; Determination of —. D. Dickinson, 409.
- Chrome Green:** Analysis of —. J. H. Van der Meulen, 133.
- Chromic Index:** Determination of —. A. Ionescu-Matiu, C. Popesco and O. Constantinesco, 135.
- Chromic Oxidation:** Volumetric determination of organic substances by —: Nitrochromic solutions. M. H. Cordebard, 123.
- Chromic Oxide** in leather; Determination of —. J. C. Mertz, 310.
- Chromium** in chrome-tanned leather; Determination of —. R. M. Lollar, 531.
Isotopes of —. 568.
- Chutney:** "Sweet mango" —. 291.
- Cider:** Lead in —. 28.
- Cinnamon:** Quantitative determination of powdered —. A. H. Saber, 421.
Quinine and — tablets. 29.
- Citrus juices;** Gases in the commercial handling of —. G. N. Pulley and H. W. von Loesecke, 42.
trees; Effect of fluorine insecticides on —. 229.
- Clay** products; Effect of lime in fired —. 598.
- Clays:** Adsorption of malachite green by —. V. L. Bosazza, 670.
Determination of silicic acid in —. 132.
- Clover nodule bacteria.** 105.
- Coal(s):** Analysis of commercial grades of —. Part II. 32.
Application of rational analysis to some typical —. G. K. Morrison, 376.
dust; Precautions against —. 293.
Extraction of — with quinoline. R. Belcher and R. V. Wheeler, 614.
- Methods of analysis of —. Fuel Research Board Report No. 44. 648.
- *Net calorific value of —. T. Evans, 353.
- Coal Mines Act, 1911:** Precautions against coal dust. 293.
- Coal Tar** naphtha distillates; Toxicity of —. H. Taylor, 121.
- Coatings** for protection of foodstuffs; Gelatinous —. 599.
- *Cobalt:** Cyanometric determination of nickel and —. W. J. Agnew, 643.
deficiencies in soil. 105.
deficiency in pastures. 33.
in animal organs. 34.
in foods and biological materials; Bibliography on —. 513.
in foods; Determination of —. N. D. Sylvester and L. H. Lampitt, 424.
Non-precipitation of — by nitro- β -naphthol. C. Mahr and W. Prodingier, 187.
- Cocaine:** Colour reaction for identifying —. M. Pesetz, 48.
Toxicological detection of —. A. Brüning, 427.
- *Cocoa** alkaloids in confectionery; Determination of —. J. C. Harral, 408.
- Coconut Oil:** fatty acids in Chinese —; Composition of. H. Nobori, 658.
Thermo-stability of —. 301.
- Coffee:** Composition of "special" types of —. 165.
- Coke:** Methods of analysis of —. Fuel Research Board Paper No. 44. 648.
- Colloid Chemistry—**a Textbook. (Review), H. B. Weiser, 536.
- Colne:** Appointment of F. Maudsley as Borough Analyst for —. 161.
- Compost:** its preparation and use. E. R. Orchard, 416.
- Compounds:** Organic —. *See Organic.*
Piperidyl derivatives of aromatic halogenonitr —; Identification of. M. K. Seikel, 530.
Rules for naming inorganic —. 509.
- Concrete** products; "Crazing" of cast —. 598.
Resistance of — to attack by chemical agents. 598.
- Congo Red** test for examining damaged cotton. Further developments and applications. G. G. Clegg, 475.
- Contraphthisine:** A reputed cure for tuberculosis. 48.
- Cooling Baths:** Low-temperature —. H. L. Wikoff, B. R. Cohen and M. H. Grossman, 383.
- Copper:** Colorimetric determination of — with triethanolamine. J. H. Yoe and C. J. Barton, 578.
Determination of — with the aid of the silver reductor. N. Birnbaum and S. M. Edmonds, 313.
in apple leaves. 292.
in butter; Determination of —. G. M. Moir and E. D. Andrews, 657.
in concentrated tomato pulp. 104.
*in presence of cadmium; Electrolytic determination of small quantities of —. J. G. Fife, 562.

Copper—*continued*

- *in tomato pulp, 28; C. A. Mitchell, 220.
- in tomatoes. W. J. Shannon and D. T. Englis, 605.
- in tomatoes and tomato purée, 28.
- *Potentiometric titration of glucose with alkaline tartrate solutions of —. H. T. S. Britton and L. Phillips, 18.
- Reaction of — with benzidine. G. Spacu and C. G. Macarovici, 248.
- Sensitive reaction for —. G. H. Wagenaar, 533.
- *Separation of — from cadmium with quinaldinic acid as reagent. A. J. Lindsey and R. J. Shennan, 636.
- zinc in presence of —; Micro-determination of, with quinaldinic acid as a reagent. P. R. Ráy and T. C. Sarkar, 435.

Cordials: silver in —; Estimation of. O. Noetzel, 364.

Cotton: Examination of damaged — by the Congo red test. G. G. Clegg, 475.

Pink staining of bleached —. 228.

Cotton Wool: Composition of —. 228.

Cottonseed Oil: Low temperature crystallisation of —. T. P. Hilditch and L. Maddison, 656.

Couldson and Purley: Appointment of E. Hinks as Public Analyst and D. D. Moir as Deputy Public Analyst for Urban District of —. 161.

Cowpeas: Vitamin A activity and vitamin B₁ content of —. J. O. Halverson and F. W. Sherwood, 470.

Crab meat: Colon bacilli in —. 227.

Crabs: Fat of land — in Seychelles Islands. T. P. Hilditch and K. S. Murti, 518.

Crayons: Lead in —. 164.

Cream: Alkaline —. 651.

Cheese. *See* **Cheese**.

Pasteurisation of — by the "flash" method. 600.

whipped —; Nitrous oxide for. 651.

Creatinine: Determination of — with *m*-dinitrobenzoic acid. E. Komm and H. Pinder, 229.

Relation of cuprous — to tests for sugar in urine. M. Samson, 55.

Crewe: Appointment of T. R. Hodgson as Borough Analyst for —. 161.

Crops and micro-organisms. 105.

Crosby: Appointment of W. H. Roberts as Public Analyst for Borough of —. 411.

***Crucibles**: Adjustable support for —. H. C. Lockwood, 160.

B.S.I. standard dimensions for silica —. 41.

Cryolite: silica in —; Determination of. J. Spielhacsek, 315.

silicon in —; Determination of. T. Nakamura, 434.

***Cryoscope**: Modification of stirring device of the Hortvet —. W. J. Blackie and G. F. Flemons, 26.

Crystal Chemistry: Introduction to —. (Review), R. C. Evans, 75.

Cubé: deguelin in —; Determination of. L. D. Goodhue and H. L. Haller, 184.

Cupferron: Elimination of iron by — prior to colorimetric determination of lead with dithizone. L. Panousse-Digeaud and H. Cheftel, 130.

Cupric Ammonium Chloride as means of determining aluminium oxide in metallic aluminium. T. Nakamura and S. Yamazaki, 68.

Cuprous Chloride: Precipitation of alkaloids with —. J. J. L. Zwikker and A. Krusse, 173.

Cuprous Iodide reagent; Action on alkaloids: Precipitation and colour reactions. M. Péronnet and J. Guénin, 301.

Currents: Paraffin oil on —. 164.

Currency Notes: Examination of burnt —. 166.

Curry Powder: Lead in —. 602.

Cyanide: Argentometric determination of — with diphenylcarbazon as indicator. R. R. Ripan-Tilici, 315.

in brass electroplating baths; Determination of free —. W. M. McNabb and S. Heiman, 315.

Cyanides in presence of thiocyanates and chlorides; Determination of —. A. Slooff and D. Van Duyn, 250.

***Cyanometric** determination of cobalt and nickel. W. J. Agnew, 643.

D

***Damsons**: Composition of —. 550, 554.

Daphnia method of evaluating the activity of powders of *Veratrum viride*. I. Cohen, 184.

Dartford: Appointment of F. W. F. Arnaud as Borough Analyst for —. 161.

Darwen: Appointment of J. R. Stubbs as Public Analyst and A. Lees as Deputy Analyst for Borough of —. 411.

Datura poisoning. 36.

Deguelin in derris and cubé; Determination of —. L. D. Goodhue and H. L. Haller, 184.

Denigés Reagent: Modified — for determining methylpropene. A. Newton and E. H. Buckler, 473.

Depilatories: sulphides in —; Determination of. E. M. Hoshall, 579.

thioglycolates in —; Determination of. E. M. Hoshall, 667.

Derris: deguelin in —; Determination of. L. D. Goodhue and H. L. Haller, 184.

Derris malaccensis: Isolation of malaccol from —. S. H. Harper, 472.

Derris Root constituents; New derivatives of —. T. M. Meyer and D. R. Koolhaas, 244.

rotenone in —; Determination of. T. M. Meijer and D. R. Koolhaas, 526.

***Desiccating agent**: Use of anhydrite as —. H. Hawley, 644.

Deuterium for investigating the inertia of highly unsaturated fatty acids in the animal. K. Bernhardt and R. Schoenheimer, 466.

Diabetic chocolate; *d*-Sorbitol in —. 651.

Diacetyl: Colorimetric estimation of —. E. Komm and J. Flügel, 361.

Dialyser: Rapid circulating —. A. R. Taylor, A. K. Parpart and R. Ballentine, 253.

2,4-Diaminodiphenylamine: Separation and determination of —. I. S. Shupe, 666.

Diarylamines: New colour reaction for —. E. M. Meade, 530.

***2,2'-Dichlorodiethyl Sulphide**: Reaction between bleaching powder and —. A. G. Lipscomb, 100.

2,6-Dichlorophenol-Indophenol solutions; Preparation of stable —. I. Stone, 660.

Dictionary: Chambers's Technical —. (Review), C. F. Tweeney and L. E. C. Hughes, 622.

Dielectrics: Research on —. 414.

Diethylene Glycol in essence of lemon. 651.

7,4'-Dihydroxyisoflavan in mare's urine; Colour reaction for detecting —. W. Discherl, 368.

Dimethylaminoazobenzene: Effect of oral administration of — on growth of rats. J. White, 467.

Dimethylglyoxime: Titration of nickel with — with the aid of the dropping mercury electrode as indicator electrode. I. M. Kolthoff and A. Langer, 476.

m-Dinitrobenzoic Acid as means of determining creatinine. E. Komm and H. Pinder, 229.

3,5-Dinitro-4-Methylbenzazide as a reagent for identifying amines. P. P. T. Sah, 125.

Diphenylcarbazide as means for colorimetric determination of lead as chromate. T. V. Letonoff and J. G. Reinhold, 433.

Diphenylcarbazone as indicator in the argentometric determination of cyanide. R. Ripan-Tilici, 315.

***Dishes**: Adjustable support for —. H. C. Lockwood, 160.

Petri —. B.S.I. specification for. 463.

***Dithiol** as means of determining tin in food-stuffs. R. de Giacomi, 216.

Dithizone as means for colorimetric determination of small amounts of lead. H. Fischer and G. Leopoldi, 432.

colorimetric determination of lead with —; Elimination of iron by cupferron prior to. L. Panousse-Digeaud, 130.

Dog Liver Oil: 231.

Dripping (Maximum Prices) Order, 1940. 567; Fees under —. 539.

Dropsy: Epidemic — and *Argemone mexicana* oil. 226.

Drug(s): Bell's Sale of Food and —. 10th Ed. (Review), R. A. Robinson and R. Ives, 385.

Is carboic acid a —? (Legal Notes), 221.

New Zealand Pharmacy Act definition of —. 166.

*sulphanilamide group of —; Determination of members of. G. V. James, 206.

See also Food and Drugs.

Drying agent; Lead-sodium alloy as —. H. Soroos, 191.

Duboisia myoporoides: New solanaceous alkaloids from —. W. Mitchell, 233, 422.

Dust(s) from lungs produced by mining and other processes; Examination of —. D. G. Beadle, 472.

Dyed materials; Machine for testing the fastness to rubbing of —. D. A. Derrett-Smith and H. B. Bradley, 534.

Dyes: Absorption of substantive — by oxy-celluloses of the acidic type. S. M. Neale and W. A. Stringfellow, 247.

artificial — in tomato pulp; Detection of. H. Thaler and K. E. Schulte, 300.

Hair —. II. Functions and reactions of phenols. H. E. Cox, 393.

Dyestuffs: Identification of groups of — by their reaction with ferrous hydroxide. H. Eichler, 431.

E

Eccles: Appointment of G. H. Walker as Public Analyst for Borough of —. 27.

Eczema: Facial — in New Zealand. 34.

Effluents: free chlorine in —; Colorimetric determination of. F. J. Hallinan, 578.

from milk factory. 102.

*sewage —; Rapid stability test for. D. Dickinson and E. M. Weir, 290.

Egg albumin; Determination of reducing groups with porphyrindin, with special reference to —. E. Brand and B. Kassell, 425.

albumin; Molecular weight of —. F. W. Bernhart, 236.

-white injury; Attempts to isolate the factor (vitamin H) curative of —. P. György, R. Kuhn and E. Lederer, 117.

-white injury; Curative factor (vitamin H) for —, with particular reference to its presence in different foodstuffs and in yeast. P. György, 117.

-white injury; Physicochemical properties of the factor (vitamin H) curative of —. T. W. Birch and P. György, 117.

*yolk; Determination of carotene in —. 277.

yolk; Presence of *l*-tyrosine in alcoholic extract of —. L. Bracaloni, 298.

yolk; Vitamin A, carotene and xanthophyll content of —. B. Sjollem and W. F. Donath, 521.

Egyptian hashish; Isolation of cannabidiol from —. A. Jacob and A. R. Todd, 466.

*marking ink; An ancient —. C. A. Mitchell, 100.

Ehrlich's Reaction: Indole determination by a modification of —. L. H. Chernoff, 465.

Eire: Report of State Chemist for 1939-40. T. S. Wheeler, 459.

Electrical standards. 414.

Electrochemical Analysis; Electrochemistry and —. Vol. II (Review), H. J. S. Sand, 620.

Electrochemistry and Electrochemical Analysis. Vol. II. (Review), H. J. S. Sand, 620.

Theoretical and Applied —. 3rd Ed. (Review), M. de K. Thompson, 193.

Electrode: Dropping mercury — as indicator — in the titration of nickel with dimethylglyoxime. I. M. Kolthoff and A. Langer, 476.

***Electro-Deposition** of tin from chloride solutions; Quantitative —. F. G. Kny-Jones, A. J. Lindsey and A. C. Penney, 498.

- *Electrodialysis** in the analysis of soils; Observations on use of —. M. Puffeles, 501.
 New cell for —, especially of soils. A. W. Marsden, 435.
 of soils. Influence of exchangeable bases on the recovery of manganese by —. R. C. Hoon and C. L. Dhawan, 526.
- *Electrolysis**: Studies in interval —. V. Determination of small quantities of copper in presence of cadmium. J. G. Fife, 562.
- Electrolytic Analysis** and — Marsh tests. (Review), H. J. S. Sand, 620.
- Electrophoretic Fractionation** as means of characterising starch from various sources. O. Dahl, 362.
- Electroplating** baths; Determination of free cyanide in brass —. W. M. McNabb and S. Heiman, 315.
- Elements**: Atomic weights of —. 415.
 Discovery of the —. 4th Ed., (Review), M. E. Weeks, 321.
 in organic compounds; Detection of —. L. Rosenthaler, 307.
 *in organic compounds; Micro-tests for —. II. Phosphorus, arsenic and antimony. C. L. Wilson, 405.
 Luminescence analysis of rare —. M. Servigne, 382.
- Elephant Liver Oil**: 231.
- Emodic Acid**: a metabolic product of *Penicillium cyclopium* Westling. W. K. Anslow, J. Breen and H. Raistrick, 470.
- Engineering Science**; Records and Research in —. (Review), J. E. Holmstrom, 322.
- Enzyme activity** in frozen vegetables: Asparagus. M. A. Joslyn and C. L. Bedford, 468.
- Enzymes** of gum arabic; Inactivation of —. J. P. Kleff, 112.
- Equol** of mare's urine; Colour reaction for detecting —. W. Discherl, 368.
- Ergometrine**: Colorimetric determination of —. S. A. Schou and J. Bennekou, 51.
- Ergosterol**: Production of — by *Penicillium carmino-violaceum*. H. G. Hind, 470.
- Ergot**: Chemical assay of powdered —. C. Daglish and F. Wokes, 50.
- Ergotoxine**: Colorimetric determination of —. S. A. Schou and J. Bennekou, 51.
- Erith**: Appointment of F. W. F. Arnaud as Public Analyst for the Borough of —. 27.
- Erythroglauin**: Molecular constitution of —. W. K. Anslow and H. Raistrick, 574.
- Esher Urban District**: Appointment of E. Hinks as Public Analyst and D. D. Moir as Deputy Public Analyst for —. 27.
- Essences**: Vehicle for —. 599.
- Essential Oil(s)**: eugenol content of —; Determination of. P. A. Rowaan and J. A. Insinger, 66.
 of massoi bark. T. H. Meijer, 375.
- Ether**: Anaesthetic —. B.P. Sub-Committee recommendation. 167.
 water in —; Determination of. R. Gaspari and G. Serrure, 73.
- Ethol Alcohol** in pharmaceutical products; Microdetermination of —. Determination of chromic index. A. Ionescu-Matui, C. Popesco and O. Constantinesco, 135.
- Ethylene Glycol** in lubricating oil; Detection and determination of —. H. Lamprey, E. E. Sommer and A. D. Kiffer, 665.
- Eucalyptus ointment**. H. Brindle, 608.
- Eugenol** content of essential oils; Determination of —. P. A. Rowaan and J. A. Insinger, 66.
- Europium**: Isotopes of —. 568.
- Extractum hyoscyami**: Alkaloid content of —. J. A. C. Van Pinxteren, 172.

F

- Fabrics**: Stains and defects in —. 228.
- Fat**: Coupled oxidation of carotene and — by carotene oxidase. J. B. and R. J. Sumner, 571.
 of land crabs (Seychelles Island). T. P. Hilditch and K. S. Murti, 518.
 spoilage; Chemistry of —. Analytical differentiation of aldehydes formed. K. Täufel and K. Klentsch, 44.
 Total Solids and Moisture. (Review), R. D. Mason, 75.
- Fats**: Analysis of commercial —. Report of American Chemical Society Committee, 508.
 Arylation of —. Synthesis of tolyl stearic acid and its esters. W. Kimura and J. Tsurugi, 186.
 Body — of the pig. V. Component glycerides of perinephretic and outer back — from the same animal. T. P. Hilditch and W. H. Pedelty, 569.
 Chemical Composition of Natural —. (Review), 319.
 *Determination of — by the Grossfeld method. W. H. Kitto, 97.
 flow and drop points of —; B.S.I. specification for determining. 418.
 from seeds of *Garcinia indica* (kokum butter); Fatty acids and glycerides of —. N. L. Vidyarthi and C. J. Dasa Rao, 171.
 Indian ox depot —; Component acids and glycerides of. T. P. Hilditch and K. S. Murti, 655.
 liquid seed —; Mixed unsaturated glycerides of. I. Some non-drying oils. B. G. Gunde and T. P. Hilditch, 364. II. Low temperature crystallisation of cottonseed oil. T. P. Hilditch and L. Maddison, 656.
 seed — of *Allanblackia floribunda* and *Allanblackia parviflora*; Fatty acids and glycerides of. M. L. Meara and Y. A. H. Zaky, 517.
 solid seed —; Fatty acids and glycerides of. IX. *Mimusops Heckelii* (Baku) kernel fat. D. Atherton and M. L. Meara, 517.
 Thermo-stability of —. E. Glimm, H. Wittmeyer and W. Jahn-Held, 301.
- Fatty Acid(s)** dehydrogenase IV. Product of the dehydrogenation of stearic acid. O. St. A. K. Lang and F. Adickes, 241.
 dehydrogenase and its co-enzyme. O. St. A. K. Lang and H. Mayer, 241.
 Gradual decomposition by oxidation of — into their next lower homologues. H. Mendel and J. Coops, 246.
 in Chinese coconut oil; Composition. H. Nobori, 658.
 in halibut intestinal oil; Distribution of —. Note on free — in intestines of fish. J. A. Lovern and R. A. Morton, 58.
 in herring oil; Lower saturated —. H. Nobori, 607.

Fatty Acids—continued

- Inertia of highly unsaturated — in the animal, investigated with deuterium. K. Bernhardt and R. Schoenheimer, 466.
of fat from seeds of *Garcinia indica* (kokum butter). N. L. Vidyarthi and C. J. Dasa Rao, 171.
of oil from sapota seeds. N. L. Vidyarthi and V. Mallya, 171.
of seed fats of *Allenblackia floribunda* and *Allanblackia parviflora*. M. L. Meara and Y. A. H. Zaky, 517.
of solid seed fats. IX. *Mimusops Heckelii* (Baku) kernal fat. D. Atherton and M. L. Meara, 517.
of some *Myristica* fats. D. Atherton and M. L. Meara, 517.
Separation of hydroxy from non-hydroxy — by means of a dibasic acid anhydride. F. E. Kurtz and P. S. Schaffer, 528.
Fatty Alcohols: Unsaponifiable matter in —. D. Burton and G. F. Robertshaw, 615.
Fatty Esters: Purification of high molecular weight —. L. O. Buxton and R. Kapp, 528.
Fatty Oils: Component glycerides of vegetable —. Niger seed oil. N. L. Vidyarthi and M. V. Mallya, 419.
from seeds of *Bauhinia variegata*. S. V. Puntambeka and S. Krishna, 421.
Federated Malay States: Rat virus enquiry report. 460.
Report of Institute of Medical Research for 1938. A. N. Kingsbury, 459.
***Feeding Stuffs**: Carotene and allied substances in —. 263.
Compound and Mixed — (Control) (No. 2) Order, 1940, 505. Amendment Order, 645.
*for poultry; Phytic acid content of —. R. H. Common, 79.
(Maximum Price) Order, 1940. 504.
Registration of — in South Africa. 229.
***Fehling's Solution**: Potentiometric titration of glucose with —. H. T. S. Britton and L. Phillips, 18.
Felspar: quartz particles in —; Optical method for determining. G. H. McIntyre and M. Bozain, 535.
Fermentation process in tea manufacture. IV. Tea tannin and its — products. C. J. Harrison and E. A. H. Roberts, 46; V. Cytochrome oxidase and its probable rôle. VI. Effect of dilution on rate and extent of oxidations in fermenting tea leaf suspensions. E. A. H. Roberts, 521.
Ferric Iron: Volumetric determination of — by means of mercurous nitrate. F. R. Bradbury and E. G. Edwards, 433.
Ferric Sulphate: Effect of — in shortening the digestion period in the Kjeldahl method. F. M. Stubblefield and E. E. Deturk, 616.
Ferrous materials: Accelerated spray test for determining the relative atmospheric corrodibility of —. T. Swinden and W. W. Stevenson, 670.
Ferrous Hydroxide: Identification of groups of dyestuffs by their reaction with —. H. Eichler, 431.
Fertilisers: Problems in vegetable production. E. J. Greenstein, 415.
Raw materials (—) Order, 1940. 648.
Registration of — in South Africa. 229.

- Fertilisers and Feeding Stuffs Act, 1926**: Appointments under —. 411.
***Ferula allitacea* fruit**; Oil from —. P. K. Bose and S. N. Dutt, 421.
Filter Paper for analysing tan liquors; Use of cheap —. H. B. Merrill and G. C. Mueller, 476.
***Filters**: Jena glass —; Cleaning of. H. Hawley, 27.
Testing of air —. 599.
Fire-proofing compounds; Action of — on wood. 163.
resistance of timber. 163.
Fischer volumetric method for determining water. E. G. Almy, W. C. Griffin and S. C. Wilcox, 616.
Fish: Canned —. See Canned.
fatty acids in intestines of —; Presence of. J. A. Lovern and R. A. Morton, 58.
freshness of —; Estimation of. 224.
"Pink" spoilage of salted —. 224.
Fish Liver Oils: Vitamin A destruction in —. E. J. Simons, L. O. Buxton and H. B. Colman, 469.
Fish Oils: New Zealand —. III. Composition of depôt fats of the ling (*Genypterus blacodes*). F. B. Shorland, 172.
Vitamin A content of —. 225.
Fish-Poison Plants: Active principles of leguminous —. IV. Isolation of malaccol from *Derris malaccensis*. S. H. Harper, 472.
Flash method of pasteurising cream. 600.
Flavanic Acid as means of estimating arginine. H. B. Vickery, 236.
Flavone derivatives; Boric acid colour reaction of —. C. W. Wilson, 66.
Flour: bleaching of — with chlorine; Detection of. D. B. Scott, 605.
*Carotene determined in —. 277.
self-raising —; Baking powder and. 650.
self-raising —; Double-strength. 503.
Soya-bean — in sausage estimated by determining non-fermentable sugars. W. B. Hendrey, 109.
sugars in —; Determination of. W. Iwanowski and G. Grabouska, 168.
Fluorescence analysis in the estimation of *o*-nitrophenol in *p*-nitrophenol, and *o*-aminophenol in *p*-aminophenol. W. Seaman, A. R. Norton and O. E. Sundberg, 581.
Analysis in Ultra-Violet Light. 3rd Ed. (Review), J. A. Radley and J. Grant, 194.
analysis of human urine. W. Koschra, S. von der Seipen and P. A. Aldred, 242.
of ghee in ultra-violet light as means of detecting adulteration. G. Narasimhamurthy and V. V. Suryanarayanamurthy, 606.
Fluoride: Amperometric titration of — with thorium nitrate. A. Langer, 669.
Fluorine in the hot-springs of Gastein; Determination of —. R. Bisanz and F. Kroupa, 189.
in wood treated with fluoride preservative; Determination of —. B. Ikert, 190.
insecticides; Effect of — on citrus trees, 229.
silica in materials containing —; Determination of. H. Spielhaccek, 315.
Thorium nitrate micro-titration of — in aqueous and alcoholic systems. J. W. Hammond and W. H. MacIntire, 618.

- Fluorspar:** silicon in —; Determination of. T. Nakamura, 434.
- Food and Drugs analysis.** Abstracts: 42, 108, 168, 229, 297, 360, 418, 464, 516, 569, 605, 655.
- Food and Drugs Act, 1938:** Application to Scotland. 39; to Northern Ireland, 40.
- Food Investigation Board Reports:** Function of nitrate, nitrite and bacteria in the curing of bacon and hams. Special Report No. 49. 355.
- Report for 1938. 223.
- Food(s):** Action of mustard gas on —. A. Hasskó, 427.
- Bell's Sale of — and Drugs. 10th Ed. (Review), R. A. Robinson and R. Ives, 385.
- *Carotene and allied substances in —. 263.
- Chemical composition of —. R. A. McCance and E. M. Widdowson, 458.
- cobalt in —; Determination of. N. D. Sylvester and L. H. Lampitt, 424.
- (Compound and Mixed Feeding Stuff) Order 1940. 645.
- Distribution of aneurin in — determined by chemical analysis. M. Pyke, 180.
- frozen—; Factors affecting the efficiency of materials used in packaging —. W. Rabak, 658.
- hawkers; Detention of —. 601.
- Manufacture; Handbook of —. (Review), F. Fiene and S. Blumenthal, 255.
- metals in —; Bibliography on. I. Cobalt, 513; II. Nickel, 603.
- Preservatives in —. Public Health Amendment Regulations, 1940. 359.
- standards; Report on —. 103.
- substances containing phosphorus; Estimation of vitamin D in —. K. H. Coward and E. W. Kassner, 523.
- Sulphur content of —. M. Masters and R. A. McCance, 42.
- Values at a Glance. (Review), V. G. Plimmer, 388.
- Foodstuffs:** bromine in —; Micro-determination of. W. P. Ford and Others, 617.
- Carboxide as fumigant for —. 599.
- Combined ascorbic acid in plant —. J. C. Pal and B. C. Guha, 522.
- formic acid in —; Determination of. J. Grossfeld and R. Payfer, 168.
- Gelatinous protective coating for —. 599.
- nicotinic acid in certain —; Estimation of. E. Kodicek, 522.
- Presence of curative factor (vitamin H) for egg white injury in different —. P. György, 117.
- riboflavin content of —; Fluorometric method for determining. A. Z. Hodson and L. C. Norris, 114.
- saccharin in —; Detection of. W. F. Reindollar, 569.
- *Tin in — determined by means of dithiol. R. de Giacomi, 216.
- vitamin C in —; Estimation of. P. N. Sen-Gupta and B. C. Guha, 523.
- Foot perspiration; Human — and upper boot leather.** A. Colin-Russ, 659.
- Forensic chemistry.** Abstracts: 471.
- Chemistry. (Review), H. T. F. Rhodes, 621.
- Forest Products Research Board: Report for 1938.** 163.
- *Formaldehyde in air; Determination of small amounts of —. R. W. Kersey, J. R. Maddocks and T. E. Johnson, 203.
- Formaldehyde** colorimetric method for manganese; Improved form of —. C. P. Sideris, 476.
- Formalin** tablets, 597.
- Formic Acid** in foodstuffs; Determination of —. J. Grossfeld and R. Payfer, 168.
- New reaction of —. T. L. Davis and W. P. Green, 528.
- poisoning. 459.
- Fruit(s):** Antiscorbic value of —. M. Olliver, 662.
- Arsenic in —. 229.
- Canned —. See Canned.
- *content of jams; Determination of —. C. L. Hinton and T. Macara, 540.
- cordials; Estimation of silver in catadynised —. O. Noetzel, 364.
- Gas storage of —. 224.
- gumming of Victoria plums. 512.
- Vitamin C content of West Australian —. 291.
- Fruit and Vegetable Preservation Research Station, Campden: Report for 1939.** F. Hirst, 511.
- Fuel** research. Analysis of commercial grades of coal, Part II. 32.
- Fumes:** Poisonous — from an arc light. 292.
- Fumigant** for foodstuffs; Carboxide as —. 599.
- Fumurate** in animal tissues; Determination of —. H. A. Krebs, D. H. Smyth and E. A. Evans, 571.
- Fungi:** wood-destroying —; Physiological studies of. 164.
- Furfural:** Photometric estimation of —. R. A. Stillings and B. L. Browning, 663.

G

- Galacturonic Acid:** Micro-estimation of —. E. M. Kapp, 580.
- Gallie Acid** as means for colorimetric determination of ferric iron. Y. Volmar and A. Wagner, 249.
- Gallium:** Reaction of — with perchloric acid. L. S. Foster, 68.
- Gambir:** catechin and catechin-tans in —; Determination of. C. J. van Hulssen and D. R. Koolhaas, 127.
- Garcinia indica** seeds; Fatty acids and glycerides of fat from —. N. L. Vidyarthi and C. J. Dasa Rao, 171.
- Gas analysis.** Abstracts: 244, 473, 525, 575.
- *detection apparatus for qualitative micro-chemistry. C. L. Wilson, 407.
- storage of fruit and vegetables. 224.
- Gases:** in the commercial handling of citrus juices. G. N. Pulley and H. W. von Loesecke, 42.
- toxic — in industry; Methods for detecting. Aniline vapour. D.S.I.R. Leaflet No. 11. 162; Carbon monoxide. Leaflet No. 7, 223. Phosgene. Leaflet No. 8, 290; Arsine. Leaflet No. 9, 354.

- Gastein** hot springs; Determination of fluorine and silica in —. R. Bisanz and F. Kroupa, 189.
- Geelbek**: vitamin A content of visceral organs of —; Seasonal variations in. C. J. Molteno and W. S. Rapson, 59.
- Gelatin**: Preservatives for preparations containing —. L. Gershenfeld and D. Perlstein, 62.
- General Medical Council**: Second addendum to British Pharmacopoeia, 1932. 457.
- Genypterus blacodes**: Composition of depôt fats of the ling (—). F. B. Shorland, 172.
- Germanium** detected and determined with hydrogen selenide. V. J. Kouznecov, 68.
- Ghee**: Adulteration in — detected by a study of its fluorescence. G. Narasimhamurty and V. V. Suryanarayanamurty, 606.
- Adulteration of —. 652.
- Neutralisation value of —. H. Hawley, 606.
- Sterol acetate test for —. 601.
- Gillingham**: Appointment of F. W. F. Arnaud as Public Analyst for Borough of —. 411.
- *Glass** filters; Cleaning Jena —. H. Hawley, 27.
- Glasses**: Organic and inorganic —. H. Moore, 126.
- Glucose**: Blood sugar levels in rats receiving xylose and —. W. J. Darby and P. L. Day, 520.
- *Potentiometric estimation of — with potassium ferricyanide in sodium carbonate solution. H. T. S. Britton and L. Phillips, 149.
- *Potentiometric titration of — with alkaline tartrate solutions of copper, including Fehling's solution. H. T. S. Britton and L. Phillips, 18.
- Glutamic Acid** of normal and malignant tissue proteins. A. C. Chibnall and Others, 520.
- Glycerides** of fat from seeds of *Garcinia indica* (kokum butter). N. L. Vidyarthi and C. J. Dasa Rao, 171.
- of liquid seed fats; Mixed unsaturated —. Some "non-drying" oils. I. B. G. Gunde and T. P. Hilditch, 364. II. Low temperature crystallisation of cottonseed oil. T. P. Hilditch and L. Maddison, 656.
- of oil from sapota seeds. N. L. Vidyarthi and V. Mallya, 171.
- of palm oils; Partial separation of palm oils by crystallisation as an aid to determination of —. T. P. Hilditch and L. Maddison, 517.
- of perinephritic and outer back fats from the same pig. T. P. Hilditch and W. H. Pedelty, 569.
- of seed fats of *Allanblackia floribunda* and *Allanblackia parviflora*. M. L. Meara and Y. A. H. Zaky, 517.
- of solid seed fats. IX. *Mimusops Heckelii* (Baku) kernel fat. D. Atherton and M. L. Meara, 516.
- of some Indian ox depôt fats; Component —. T. P. Hilditch and K. S. Murti, 655.
- of some *Myristica* fats. D. Atherton and M. L. Meara, 517.
- of typical cow milk fat; Component —. T. P. Hilditch and S. Paul, 606.
- Glycerides**—continued
of vegetable fatty oils. Niger-seed oil. N. L. Vidyarthi and M. V. Mallya, 419.
- Glycerin**: Effect of — on the distillation method for water. R. B. Trusler, 665.
- Glycerinum**: B.P. Sub-Committee recommendation. 167.
- Glycerol**: Formation of trimethylene glycol from — by *Aerobacter*. M. N. Nickelson and C. H. Werkman, 613.
- in blood; Determination of neutral fat — with periodate. L. Voris, G. Ellis and L. A. Maynard, 425.
- Glycogen** in meat. 224.
- of *Helix pomatia*. E. Baldwin and D. J. Bell, 468.
- Glyouronic Acid** and its conjugated compounds; Quantitative estimation of — by means of the naphthoresorcinol test of Tollens. W. Mozolowski, 519.
- Micro-estimation of —. E. M. Kapp, 580.
- Goats**: Milk of —. 226.
- Gold**: Detection of — by means of morpholin. L. S. Malowan, 310.
- *Gooseberries**: Composition of —. 543, 545.
- Gosport**: Appointment of R. P. Page as Public Analyst for Borough of —. 411.
- Government Analysts**: Notes from Reports of —. See **Bangalore, Ceylon, Eire, Federated Malay States, Madras, New South Wales, New Zealand, Palestine, Patiala (Punjab), Queensland, South Africa, United and Central Provinces of India, Western Australia.**
- Grass**: dried—; Commercial determination of carotene and allied pigments in —. W. M. Seaber, 266.
- Great Yarmouth**: Appointment of W. F. Greeves as Deputy Public Analyst for Borough of —. 566.
- *Greengages**: Composition of —. 549, 554.
- Grignard Reagent** for determination of hydroxyl groups. W. Fuchs, N. H. Ishler and A. G. Sandhoff, 663.
- for micro-gravimetric determination of active hydrogen. R. N. Evans, J. E. Davenport and A. J. Revukas, 533.
- prepared from magnesium amalgams. E. G. Rochow, 245.
- *Grossfeld Method** for determining fat. W. H. Kitto, 97.
- Groundnut**: Bambarra —. J. M. Holm and B. W. Marloth, 608.
- Gum Arabic**: Inactivation of enzymes of —. J. P. Kleff, 112.

H

- Haemoglobinometer** standardisation. 414.
- Haenium**: New isotope of —. 568.
- *Hair** dyes. II. Functions and reactions of phenols. H. E. Cox, 393.
- Examination of —. 38.
- Lead content of human —. K. N. Bagchi, H. D. Ganguly and J. N. Sirdar, 471.
- Halibut Intestinal Oil**: fatty acids in —; Distribution of. J. A. Lovern and R. A. Morton, 58.

- Halifax**: Appointment of R. Mallinder as Public Analyst and Agricultural Analyst for County Borough of —. 411.
- Halogenonitro** compounds; Identification of aromatic — as piperidyl derivatives. M. K. Seikel, 666.
- Hamburg Steak**: Pork in —. 227.
- Hams**: Function of nitrate, nitrite and bacteria in the curing of —. 355.
- Harrogate**: Appointment of F. W. M. Jaffé as Public Analyst for Borough of —. 161.
mineral water; Presence of sulphide in — and its oxidation by air. A. Woodmansey, 430.
- Hashish**: Application of Beam's test for —. 166.
Isolation of cannabidiol from Egyptian —. A. Jacob and A. R. Todd, 466.
- Heat** penetration in the canning of meat in the home by the pressure cooker; Effectiveness of —. C. I. Nelson and D. Berrigan, 120.
- Helix pomatia**: Glycogen of —. E. Baldwin and D. J. Bell, 468.
- Hemicelluloses** of the wood of English oak. 5. Structure of hemicellulose B. M. H. O'Dwyer, 475.
- Hen Liver Oil**: 231.
- Hera pathite** reaction applied to aristoin. M. Wagenaar, 111.
- Hereford**: Appointment of F. G. D. Chalmers as Additional Public Analyst for Borough of —. 411.
- Herring**: Significance of bacterial count in determining freshness of — and the raw materials of kippered —. E. Aschehoug and R. Vesterhus, 417.
- Herring Oil**: Lower saturated fatty acids in —. H. Nobori, 607.
Rancidity of —. 224.
- Histidine** in urine; Detection and estimation of —. E. Racker, 467.
- Holarrhena antidysenterica**, Wall. An anti-dysenteric drug, containing poisonous alkaloids. 602.
- Home Office** Statutory Rules and Orders, 1940. Nos. 452 and 453. Poisons. 412.
- Honey**: Artificial —. 166.
Iodine absorption of genuine and artificial —. C. Griebel and G. Hess, 299.
Reducing power of unripe —, and — from sugar-fed bees. R. F. Kardos, 363.
Vitamin C content of —. E. Becker and R. F. Kardos, 470.
- Hops**: preservative value of —; Colorimetric determination of. Standard colour values of some hybrid —. A. A. D. Comrie, 516.
- *Horvot** cryoscope; Modification of the stirring device in —. W. J. Blackie and G. F. Flemmons, 26.
- Huddersfield**: Appointment of R. Mallinder as Public Analyst and Agricultural Analyst for County Borough of —. 411.
- Hydnocarpus anthelmintica** Oil: Analysis of —. H. I. Cole and H. T. Cardoso, 231.
- Hydnocarpus wightiana** Oil: Analysis of —. H. I. Cole and H. T. Cardoso, 45.
and its derivatives; Keeping properties of —. U. P. Basu and A. Mazumdar, 465.
- Hydrazine** in alkaline solution; Potentiometric determination of —. H. T. S. Britton and M. Königstein, 532.
- Hydrocarbons** in unsaponifiable matter; Determination of —. J. Grossfeld, 375.
Physical Constants of —. Vol. II. (Review), G. Egloff, 585.
- Hydrocellulose**: Microchemical determination of reducing power of — (micro-copper, micro-ferricyanide and micro-iodine numbers). E. Geiger and G. Müller, 618.
- Hydrocyanic Acid**: Determination of — by the picric acid method and the KWSZ photometer. J. T. Sullivan, 122.
- Hydrogen**: Atomic weight of —. 415.
Elimination of traces of carbon monoxide from —. K. Kawakita and B. Ichinagami, 575.
Micro-gravimetric determination of active — by the Grignard reagent. R. N. Evans, J. E. Davenport and A. J. Revukas, 533.
"swells" in canned fruits. T. P. Hoar, 43.
- para-Hydrogen**: Separation of — from oxygen and carbon monoxide. E. Bergmann, L. Farkas and L. Sandler, 244.
- Hydrogen Cyanide** gas; Toxicity of flesh of rabbits killed by —. 104.
- Hydrogen ion Concentration**: Application of — to boiler water treatment. G. W. Bond, 429.
of canned vegetables. 512.
- Hydrogen Peroxide**: Removal of phosphoric acid from —. S. R. Dickman and R. H. Bray, 477.
- Hydrogen Selenide** for detecting and determining germanium V. J. Kouzнецov, 68
- Hydrogen Sulphide** in corned beef. A. Elias, 605.
Precipitation reactions of organic arsenic compounds with —. 247.
- Hydroculphite** content of indigo vats; Colorimetric estimation of —. J. Lotichius, 668.
- Hydroxy-amino-acids**: Isolation of — from protein hydrolysates. R. L. M. Synge, 238.
- 6-Hydroxychromans**: Oxidation products of α -tocopherol and of related —. L. I. Smith, W. B. Irwin and H. E. Ungnade, 115.
- ω -Hydroxyemodin**, a metabolic product of *Penicillium cyclopium* Westling. W. K. Anslow, J. Breen and H. Raistrick, 470.
- Hydroxyl Groups** determined with the Grignard reagent. W. Fuchs, N. H. Ishler and A. G. Sandhoff, 662.
Rapid qualitative test for alcoholic —. F. R. Duke and G. F. Smith, 527.
- Hydroxylamine** in alkaline solution; Potentiometric determination of —. H. T. S. Britton and M. Königstein, 532.
- 8-Hydroxyquinoline** as means for the gravimetric determination of antimony. T. I. Pirtea, 188.
as means of determining cerium. R. Berg and E. Becker, 312.
method for micro-determination of zinc. IV. C. Cimerman and P. Wenger, 381.

I

- *Ilin:** Determination of —. 212.
- Ilford:** Appointment of B. Dyer and G. Taylor as Joint Public Analysts for Borough of —. 27; of J. H. Hamence as Joint Public Analyst for Borough of —. 411.
- Incendiary Bombs:** Testing incombustible material resistant to —. B. S. I. Specification No. BS/ARP 27. 297.
- Indicator(s):** Barium rhodizonate as — in the volumetric determination of small quantities of barium and sulphate. C. C. Miller, 477. Diphenylcarbazone as — in the argentometric determination of cyanide. R. Ripan-Tilici, 315.
- Indigo:** Colorimetric estimation of —. J. Lotichius, 668. Estimation of — on the fibre. J. Lotichius, 668. vats; Colorimetric estimation of sodium hydrosulphite and the hydrosulphite content of —. J. Lotichius, 668.
- Indole** determination by a modification of Ehrlich's reaction. L. H. Chernoff, 465.
- Indo-Oxine:** a new precipitant for metals. R. Berg and E. Becker, 378.
- Industrial Science:** Records and Research in —. (Review), J. E. Holmstrom, 322.
- Infra-Red** Photography: Its Principles and Applications. (Review), W. Clark, 480.
- *Ink:** marking-; An ancient Egyptian —. C. A. Mitchell, 100. Printing — examined by ultra-violet light. 38.
- Inorganic analysis;** Abstracts: 68, 129, 187, 248, 310, 378, 432, 476, 531, 578, 616, 668. Analysis; Text-Book of Quantitative —. (Review), A. I. Vogel, 195. Chemicals; B.P. Sub-Committee on —. Recommendations, 167. Chemistry. F. Ephraim. 3rd Eng. Ed. by P. C. L. Thorne and A. M. Ward. (Review), 584. Chemistry; Mellor's Modern —. 9th Ed. (Review), G. D. Parkes and J. W. Mellor, 479. compounds; Determination of arsenic in —. D. T. Lewis and V. E. Davis, 531. compounds; Rules for naming —. 509. ions; Qualitative reactions of salicylaldoxime with —. J. F. Flagg, and N. H. Furman, 668.
- Insecticides:** Arsenical — and wine. 166. Effect of fluorine — on citrus trees. 229. Materials for plant — from Empire sources. 574.
- Insects** in wood; Destruction of — by fumigation. 164. wood-boring — detected by X-rays. 164.
- Institute of Brewing:** Report of Brewery Effluents Committee. 356.
- International Society of Leather Trades Chemists:** Determination of colour of vegetable tannin solutions. 568.
- International Union of Chemistry:** Fifth Report of Committee on Atoms, 568. Rules for naming inorganic compounds. 509. Tenth Report of Committee on Atomic Weights, 415.
- Iodate** method for determining barium; Modified —. F. C. Guthrie, 434.
- Iodide:** Detection of —. D. Hart and R. Meyrowitz, 532. in mineral waters containing bromide and large quantities of chloride; Determination of —. E. Müller and W. Stumpf, 245. Spot test for —. C. L. Wilson, 478.
- Iodine** absorption of honey and artificial honey. C. Griebel and G. Hess, 299. Colourless tincture of —. 565. determinations; Permanganate acid ashing micro-method for —. I. Values in blood of normal subjects. D. S. Riggs and E. B. Man, 579. in quinine iodobismuthate and its injectable preparations; Determination of —. G. N. Thomis and G. P. Kopanaris, 49. in soils, plant material and waters; Determination of —. G. S. Fraps and J. F. Fudge, 305. in thyroid and its preparations determined by cerate oxidimetry. W. W. Hilty and D. T. Wilson, 174.
- *Iodine Value** of soft paraffins. H. Brindle, 409.
- Ipecacuanha:** Alteration in B.P. formula for —. 457.
- Iron:** aluminium in presence of phosphoric acid and —; Determination of. G. Balanescu and M. D. Motzoc, 188. Atomic weight of —. 415. Colorimetric determination of — with salicylaldoxime. D. E. Howe and M. G. Mellon, 578. Determination of — as phosphate. 339. Elimination of — by cupferron prior to colorimetric determination of lead with dithizone. L. Panousse-Digeaud and H. Cheftel, 130. ferric-; Colorimetric determination of — by means of gallic acid. Y. Volmar and A. Wagner, 249. in aluminium and alumina; Colorimetric determination of. —. L. Roelen, 187. in bread and bread ingredients; Determination of —. C. Hoffmann, T. R. Schweitzer and G. Dalby, 569. in butter; Determination of —. G. M. Moir and E. D. Andrews, 657. Isotopes of —. 568. Limit test for —. B. P. Sub-Committee recommendation. 167. non-haemin —; Improved method for estimating. G. Brückmann and S. G. Zondek, 659. Separation of zinc from —. 381. Specific heat and melting-point of very pure —. 413. tablets; Saccharated —. 565.
- Irrigation** purposes; Standard for water for —. 292.
- Isasin:** Reactions for identifying —. M. J. Schulte, 112.
- Isotopes:** International Table of Stable —. Recommended changes in. 568.

J

- Jam**: Definition of — in Palestine. 166.
 *fruits; Composition of some — and determination of fruit content of —. C. L. Hinton and T. Macara, 540.
 *Improvements in lead precipitation method as applied to —. 556.
 (Maximum Prices) Order, 1940. 566.
 Raspberry — below fruit standard. 597.
 *Jena Glass filters; Cleaning —. H. Hawley, 27.
Jungle Potato poisoning. 36.

K

- Keighley Borough**: Appointment of F. W. M. Jaffé as Public Analyst for —. 27.
Kendal: Appointment of W. H. Roberts as Public Analyst for Borough of —. 411, 456.
Kent: Report of County Analyst for Fourth Quarter 1939, 221.
Kesteven (Linca.): Appointment of W. W. Taylor as Public Analyst for —. 27.
Ketones: Colorimetric assay of weakly phenolic —, "oestrone," in extracts of human urine. N. B. Talbot and Others, 609.
Kingston-upon-Hull: Appointment of D. J. T. Bagnall as Public Analyst for County Borough of —. 566.
Kingston-upon-Thames: Appointment of E. Hinks as Public Analyst for Borough of —. 411.
Kjeldahl method; Effect of ferric sulphate in shortening the digestion period in —. F. M. Stubblefield and E. E. Deturk, 616.
Kober Method for determining total oestrogens in a mixture of oestrogenic steroids. Modification of —. C. Bachman, 118.
Kokum Butter: Fatty acids and glycerides of —. N. L. Vidyarthi and C. J. Dasa Rao, 171.

L

- Lao** in glazed candies; Volumetric estimation of —. N. M. Molnar and J. Grumer, 175.
Lactic Acid: Determination of — by means of periodic acid. R. Boisson, 473.
 ***Lard**: beef fat in —; Detection of. K. A. Williams, 596; Erratum, 645.
 *Examination of —. R. W. Sutton and Others, 623.
 Thermo-stability of —. 301.
 tristearin in —; Detection of. 508, 596.
Larocaine: Detection of —. 173.
Lavolnier. (Review), J. A. Cochrane, 320.
 ***Lead alloys**; Simple test for arsenic in —. R. G. Robinson, 159.
 Biochemical behaviour of —. I. Influence of calcium, phosphorus, and vitamin D on — in blood and bone. A. E. Sobel and Others, 235.
 Colorimetric determination of — as chromate with the aid of diphenylcarbazine. T. V. Letonoff and J. G. Reinhold, 433.
 colorimetric determination of — with dithizone; Elimination of iron by cupferron

Lead—continued

- prior to. L. Panousse-Digeaud and H. Cheftel, 130.
 Colorimetric determination of small amounts of — by means of dithizone. H. Fischer and G. Leopoldi, 432.
 compounds in cellulose spray paints. 292.
 content of fresh vegetables; Limit for —. 291.
 content of human hair. K. N. Bagchi, H. D. Ganguly and J. N. Sirdar, 471.
 in beer and cider. 28.
 in bone. 165.
 in canned fish. 28.
 in canned food. 459.
 in crayons. 164.
 in curry powder. 602.
 *in drinking water, 102; Estimation of —. C. H. Manley, 403.
 in samsu. 459.
 in tin coatings; Determination of —. P. Foerster, 476.
 in urine; Determination of —. 291.
 Industrial poisoning with —. 291.
 *precipitation method; Improvements in — as applied to jam. 556.
 Quantitative determination of —. B. P. Sub-Committee recommendations. 167.
 Spot tests for —. E. A. Kocsis, 190.
Lead Arsenate and its spray residues; Toxicity to sheep of —. J. L. St. John and Others, 575.
 Chemical effect on — of certain salts present in soil and spray waters. J. M. Ginsberg, 613.
Lead Dioxide: Volumetric determination of —. C. Mahr and H. Ohle, 433.
Lead-Sodium Alloy as a drying agent. H. Soroos, 191.
Leather: chromic oxide in —; Determination of. J. C. Mertz, 310.
 chromium in chrome-tanned —; Determination of. R. M. Lollar, 531.
 contaminated with mustard gas; Decontamination of —. 39.
 Upper boot — and human foot perspiration. A. Colin-Russ, 659.
Legal Notes: 30, 221, 565.
Leigh (Lancs.): Appointment of J. R. Stubbs as Public Analyst for Borough of —. 27.
Lemon essence; Dimethylene glycol in —. 651.
Lewisite: Reactions of Nessler's reagent with —. J. Delga, 246.
Lignin in tannin materials; Estimation of —. J. G. Shrikhande, 531.
Lime in fired clay products; Effect of —. 598.
Ling: Composition of depot fats of —. F. B. Shorland, 172.
 ***Linoleic Acid** and its glycerides; Use of thiocyanogen values in determining —. T. P. Hilditch and K. S. Murti, 437.
 ***Linolenic Acid** and its glycerides; Use of thiocyanogen value in determining —. T. P. Hilditch and K. S. Murti, 437.
 in leaves of land plants. M. Tsujimoto, 664.
 Thiocyanogen value of —. J. P. Kass and Others, 528.
Liquid Air: Respiration of animal tissue after freezing in —. F. Lynen, 425.

- Lithium**: Isotopes of —. 568.
Quantitative analysis of plant tissues for — by the Ramage flame spectrographic method. N. L. Kent, 581.
precipitated by uranyl acetate reagents for sodium; Composition of —. E. R. Caley and W. O. Baker, 131.
- Liver** of mammals, birds, reptiles and man; Vitamin A and carotenoids in —. H. B. Jensen and T. K. With, 179.
- Liver Oils** of some terrestrial animals. M. Tsujimoto and H. Koyanagi, 231.
- Lobster meat**; Colon bacilli in —. 227.
- Lowestoft**: Appointment of W. F. Greeves as Deputy Public Analyst for Borough of —. 566.
- Lubricating Oil**: ethylene glycol in —; Detection and determination of. H. Lamprey, E. E. Sommer and A. D. Kiffer, 665.
- Lubrication** research. 414.
- Luminescence** analysis of rare elements. M. Serigne, 382.
- Lungs**: Examination of dust from silicotic —. D. G. Beadle, 471.
- Lutecium**: Atomic weight of —. 415.
- Luximeter**: Construction and application. W. L. Carson, 534.
- Lycopene**: carotene in presence of —; Determination of. G. S. Fraps, A. R. Kemmerer and S. M. Greenberg, 608.
- Lyctus** (powder-post beetles); Control of —. 164.

M.

- *M. & B. 693**: Determination of —. 210.
- Macassar oil**: Paraffin as —. 165.
- Macatuba Oil** (Mocaya). M. Silva, 605.
- Madras**: Adulteration in —. 601.
Report of Chemical Examiner to Government of — for 1938. S. R. Naidu, 36; for 1939, S. R. Naidu, 602. Report of Government Analyst for 1938-39. H. Hawley, 601.
- Magnesium** amalgams; Preparation of Grignard reagents from —. E. G. Rochow, 245.
Colorimetric determination of —. C. P. Sideris, 478.
Detection of — by means of *p*-nitrobenzeneazoresorcinol. J. P. Mehlig and K. R. Johnson, 314.
in biological material; Determination of —. J. P. Nielsen, 176.
*in nickel alloys; Determination of —. R. C. Chirside, L. A. Dauncey and P. M. C. Proffitt, 446.
Test for —. E. Eegriwe, 248.
- *Magnesium Trisilicate**: Routine examination of —. J. L. Pinder, 90.
- Malacool**: Isolation from *Derris malaccensis*. S. H. Harper, 472.
- Malachite Green**: Adsorption of — by clays and allied materials. V. L. Bosazza, 670.
- Malate** in animal tissues; Determination of —. H. A. Krebs, D. H. Smyth and E. A. Evans, 571.
- Malden and Coombe**: Appointment of E. Hinks as Public Analyst for Borough of —. 27.

- Maleic Acid**: synthetic resins containing —; Analysis of. E. Sadolin, 128.
- Malt** extracts; Determination of amino-nitrogen in —. S. R. Snider, 464.
- Mammals**: Vitamin A and carotenoids in liver of —. H. B. Jensen and T. K. With, 179.
- Manchester** Chamber of Commerce Testing House and Laboratory. Report for 1939. 228.
- Manganese** deficiencies in soil. 104.
formaldoxime colorimetric method for —; Improved form of. C. P. Sideris, 476.
in organic material containing large amounts of calcium and chlorides; Estimation of —. T. W. Ray, 570.
in the pea seed; Distribution of — in relation to marsh spot. H. H. Glasscock and R. L. Wain, 372.
method for determining base exchange capacity of soils and other materials. C. A. Bower and E. Truog, 614.
Recovery of — from soils by electro-dialysis. R. C. Hoon and C. L. Dharwan, 526.
Separation of zinc from —. 381.
Toxicology of —. A. C. Lemos, 63.
- Manganese Dioxide**: Volumetric determination of —. C. Mahr and H. Ohle, 433.
- Manganous Uranyl Acetate**: Colorimetric micro-method for estimating sodium with —. E. Leva, 316.
- Manihot utilisima** poisoning. 36.
- Manfield**: Appointment of W. W. Taylor as Public Analyst for Borough of —. 411.
- Manure**: Town refuse used as —. 104, 221.
- *Margarine**: Spectrophotometric assay of vitamin A, with special reference to —. J. R. Edisbury, 484.
*vitamin D in —; Estimation of. N. T. Gridgeman, H. Lees and H. Wilkinson, 493.
- Margarine (Addition of Borax) Order, 1940**: 411, 413.
- *Marking Ink**: An ancient Egyptian —. C. A. Mitchell, 100.
- Marsh Spot**: Distribution of manganese in the pea seed in relation to —. H. H. Glasscock and R. L. Wain, 372.
- Marsh Tests**: Electrolytic —. (Review), H. J. S. Sand, 620.
- Massachusetts, Commonwealth of**: Report of Division of Food and Drugs Dept. of Public Health for 1938. H. C. Lythgoe, 226.
- Massol Bark**: Essential oil of —. T. H. Meijer, 375.
- Mastitis**: Bromothymol blue test for —. 35.
- Meat**: canning of — in the home by the pressure cooker; Effectiveness of heat penetration in. C. I. Nelson and D. Berrigan, 120.
content of sausages; Determination of —. 257.
Decomposition of — detected by Walkiewicz's method. H. Keller and H. Möller, 418.
extracts; Ultra-violet absorption of —. J. Schormüller, 297.
Glycogen in —. 224.
Transport of —. 224.

- Medical Practice**; British Encyclopaedia of —. Vol. 12, 139.
- Medical Research Council**; Chemical composition of foods. R. A. McCance and E. M. Widdowson, 458.
- Medical Research, National Institute for**: International standards for prolactin and for the gonadotrophic substance of pregnant mares' serum, 40.
- Meiklejohn's Method** of estimating aneurin in blood; Further modification of —. H. M. Sinclair, 182.
- Meinel Colour Test**: Behaviour of certain substituted allenes towards —. F. B. LaForge and F. Acree, Jr., 614.
- Melia composita** poisoning, 36.
- Melnick-Field Method** of estimating aneurin; Modification of —. A. D. Emmett, G. Peacock and R. A. Brown, 661.
- Menthols**: Separation and determination of isomeric —. R. T. Hall, J. H. Holcomb, Jr., and D. B. Griffin, 518.
- Mercurochrome**: mercury in —; Determination of. G. J. W. Ferrey, 607.
- Mercurous Nitrate** as means for volumetric determination of ferric iron. F. R. Bradbury and E. G. Edwards, 433.
- *Mercury**: Assay of —. J. Sandilands, 13.
in blood; Method for estimating small amounts of —. L. A. Crandall, 467.
in mercurochrome; Determination of —. G. J. W. Ferrey, 607.
reagent; Precipitation reactions of organic arsenic compounds with —. 247.
reduction method for determining pyrethrin I; Seil colour reaction in —. C. S. Sherman and R. Herzog, 373.
Separation of zinc from —. 381.
Spot tests for —. E. A. Kocsis, 190.
zinc in presence of —; Quinaldine acid as micro-reagent in determining. P. R. Ray and T. C. Sarkar, 435.
- Merton and Morden**: Appointment of E. Hinks as Public Analyst and D. D. Moir as Deputy Public Analyst for Urban District of —. 161.
- Mesulol**: the bitter principle of *Mesua ferrea*. P. Dutt, N. C. Deb, and P. K. Bose, 466.
- *Metal** plates; Detection of small amounts of proteins on surface of —. J. H. Young, 564.
- Metals** in foods and biological materials; Bibliography on —. I. Cobalt, 513; II. Nickel, 603.
Indo-oxine: a new precipitant for —. R. Berg and E. Becker, 378.
Internal strain in —. 413.
sodium in presence of other —; Determination of. E. C. Elliott, 616.
Surface finish of —. 414.
- Methionine**: Colorimetric test for —. M. X. Sullivan and T. E. McCarthy, 466.
Colour reaction of —. J. J. Kolb and G. Toennies, 114.
- p-Methylaminophenol**: Separation of *o*-aminophenol, *p*-aminophenol and —. I. S. Shupe, 667.
- Methylene Blue** in the volumetric determination of acridines. A. Bolliger, 423.
- Methylene Blue—continued**
reducing system of Palestine orange peels investigated by the Thunberg system. L. Frankenthal, 662.
- *2-Methyl-1:4-Naphthoquinone**: Examination and determination of —. J. L. Pinder and J. H. Singer, 7.
- Methylpropene** determined by a modified Denigès reagent. A. Newton and E. J. Buckler, 473.
- Metropolitan Water Board Bill**. Petition against —. 201.
- Microbiological** examination of butter. B.S.I. standard for methods of —. 359.
- Microchemical** analysis. Abstracts: 69, 134, 190, 251, 316, 379, 477, 533, 579, 617.
determination of reducing power of cellulose, oxycellulose and hydrocellulose (micro-copper, micro-ferricyanide and micro-iodine numbers). E. Geiger and G. Müller, 618.
- *Microchemistry**: Gas detection apparatus for qualitative —. C. L. Wilson, 407.
of rare earths. G. Beck, 382.
- Micro-Determination** of arsenic in urine; Colorimetric semi- —. J. V. Harispe, 54.
of bromine in foodstuffs. W. P. Ford and Others, 617.
of ethyl alcohol in pharmaceutical products. Determination of the chromic index. A. Ionescu-Matiu, C. Popesco and O. Constantinesco, 135.
of succinic acid. G. J. Goepfert, 580.
of sulphur; New method for washing and isolation of barium sulphate in —. R. Grangaud, 316.
of zinc by hydroxyquinoline method. IV. C. Cimerman and P. Wenger, 381.
Quantitative — with the use of ordinary analytical balances. A. A. Benedetti-Pichler and R. A. Paulson, 69.
- Micro-Diffusion** Analysis and Volumetric Error. (Review), E. J. Conway, 254.
- Micro-Electrolytic** determinations in small volumes. J. Donau, 134.
- Micro-Estimation** of amino acids; Semi- —. H. R. Ing and M. Bergmann, 57.
of threonine. R. J. Block and D. Bolling, 72.
- Microgram**: Use of the term —. 478.
- Micro-Gravimetric** determination of active hydrogen by the Grignard reagent. R. N. Evans, J. E. Davenport and A. J. Revukas, 533.
separation of zinc and uranium. E. Kroupa, 435.
- Micro-Method** for determining iodine in blood. D. S. Riggs and E. B. Man, 579.
for estimating sodium with manganous uranyl acetate. E. Leva, 316.
- Micro-Organisms**: Biochemistry of —. Emodic acid and ω -hydroxyemodin, metabolic products of *Penicillium cyclopium* Westling. W. K. Anslow, J. Breen and H. Raistrick, 470.
Crops and —. 105.
encountered in brewing industry; Thermal death time of —. S. S. Epstein and F. D. Snell, 524.
- Microphotometer**: Quantitative spectrographic analysis with the —. D. M. Smith, 295.

- ***Microscope slide**; Technique for preparing a standard — with special reference to chocolate. H. C. Lockwood, 97.
- ***Micro-Tests** for elements in organic compounds. II. Phosphorus, arsenic and antimony. C. L. Wilson, 405.
- Micro-Titration** of selenium. G. Wernimont and F. J. Hopkinson, 534.
with thorium nitrate of fluorine in aqueous and alcoholic systems. J. W. Hammond and W. H. McIntire, 618.
- ***Micro-Titrimetric** determination of carbon dioxide in carbonates. A. F. Colson, 638.
- Middlesex**: Appointment of Miss M. H. Pearson as Deputy Public Analyst for County of —, 27.
- Milk** and nutrition; Reports on —. Part IV. 31.
beverages; Addition of stabiliser to —. 599.
cheese. 565.
factory effluents. 102.
fat from a typical cow; Component glycerides of —. T. P. Hilditch and S. Paul, 606.
fat from the Turkish buffalo. A. Heiduschka and F. Cicekdagi, 297.
Freezing-point of —, 601; L. M. Lampert, 108.
Goats' —. 226.
malting; Standards for — in New South Wales. 651.
pasteurised —; Nutritive value of. 31.
riboflavin in —; Estimation of. K. M. Henry, J. Houston and S. K. Kon, 522.
Sale of — Regulations, 1939. Statutory Rules and Orders, 1939, No. 1417. 106.
- Milk Nutrition Committee**: Reports of —. 31.
- Millimicrogram**: Use of the term —. 478.
- Mimusops heckelii** (Baku) kernel fat. D. Atherton and M. L. Meara, 517.
- Mince**: Sulphur dioxide in —. 28.
- Mine** dusts; Official methods of analysis of —. 294.
- Mineral Waters** containing bromide and large quantities of chloride; Determination of iodide in —. E. Müller and W. Stumpf, 245.
Sulphide in Harrogate — and its oxidation by air. A. Woodmansey, 430.
Trade Year Book, 1939. (Review), 142.
- Minerals**: refractive index of non-opaque —; Immersion liquids for determining. E. P. Kaiser and W. Parrish, 137.
- Mining** process; Examination of dusts produced by — taken from lungs. D. G. Beadle, 472.
- Ministry of Agriculture and Fisheries**: Cream Cheeses. Advisory Leaflet No. 222. 456.
- Ministry of Food**: Compound and Mixed Feeding Stuffs (Control) (No. 2) Order, 1940. 505. Amendment Order, 645.
Dripping (Maximum Prices) Order, 1940. 539, 567.
Feeding Stuffs (Maximum Price Order, 1940). 504.
Food (Compound and Mixed Feeding Stuffs) Order, 1940, amending the Compound and Mixed Feeding Stuffs (Control) (No. 2) Order, 1940. 645.
Jam (Maximum Prices) Order, 1940. 566.
- Ministry of Food—continued**
Margarine (Addition of Borax) Order. 411, 413.
Raw Materials (Fertilisers) Order, 1940. 648.
Sausage (Maximum Prices) Order, 1940. 358; Amendment, 507.
- Ministry of Health**: Public Health (Preservatives, etc., in Food) Amendment Regulations, 1940. 359.
Report of Chief Medical Officer for 1938. Sir A. MacNalty, 103.
- Mitcham**: Appointment of E. Hinks as Public Analyst and D. D. Moir as Deputy Analyst for Borough of —. 411.
- Mixtures**: Separation of components of — by fractional centrifuging. A. Gourevitch, 136.
- Moisture**: Fat, Total Solids, and —. (Review), R. D. Mason, 75.
- Molecular Weight** of egg albumin. F. W. Bernhart, 236.
- Molybdate**: Reduction of — in the silver reductor. C. F. Hiskey, V. F. Springer and V. W. Meloche, 69.
- ***Molybdenum**: detection of —; New test for. J. H. Hamence, 152.
in biological materials; Determination of small quantities of —. D. Bertrand, 177.
in cast iron and steel; Determination of —. C. Sterling and W. P. Spuhr, 311.
Isotopes of —. 568.
rhenium in —; Determination of. C. F. Hiskey and V. W. Meloche, 669.
- *Separation of — from tin and from sulphur. D. A. Lambie and W. R. Schoeller, 281.
- Mononitroparaffins**: Colorimetric determination of primary —. E. W. Scott and J. F. Treon, 529.
- Morecambe and Heysham**: Appointment of J. R. Stubbs as Public Analyst for Borough of —. 411.
- Morpholin** as means of detecting gold. L. S. Malowan, 310.
- Mortar**: Resistance of — to attack by chemical agents. 598.
- Motor Oils**: Determination of sludge in used or oxidised —. C. G. Ludeman, 664.
- Mould** content of tomato products. 599.
- Mustard Gas**: Action of — on foods. A. Hasskó, 427.
Decontamination of leather contaminated with —. 39.
*Reaction between bleaching powder and —. A. G. Lipscomb, 100.
Reactions of Nessler's reagent with —. J. Delga, 246.
- Mycoderma** in yeast; Estimation of —. 62.
- Myristica** fats; Component fatty acids and glycerides of —. D. Atherton and M. L. Meara, 517.

N

- Naphtha**: coal-tar — distillates; Toxicity of. H. Taylor, 121.
- Naphthalene- β -Sulphonic Acid** as reagent for amino acids. M. Bergmann and W. H. Stein, 237.

- Naphthoresorcinol** test of Tollens; Quantitative estimation of glycuronic acid and its conjugated compounds by means of —. W. Mozolowski, 519.
- National Physical Laboratory**: Report for 1939. C. G. Darwin, 413.
- Neem Oil**: I. M. Qudrat-i-Khuda, S. K. Ghosh and A. Mukherjee, 465.
- *Neodymium**: Estimation of praseodymium and — in solution from their absorption spectra. J. N. Friend and D. A. Hall, 144.
- Nephelometric** determination of arsenic with Bougault's reagent; Permanent artificial standards for —. J. Thuret, 129.
- Nessler's Reagent**: Reactions of — with aqueous solutions of mustard gas and lewisite. J. Delga, 246.
- New South Wales**: Report of Government Analyst for 1937. S. G. Walton, 599; for 1938, 649.
- New Zealand** fish oils. III. Composition of dépôt fats of the ling (*Genypterus blacodes*) F. B. Shorland, 172.
- Pharmacy Act. Definition of a drug. 166.
- Report of Dept. of Agriculture for 1938-39. R. E. R. Grimmett, 33.
- Newton-le-Willows**: Appointment of J. R. Stubbs as Public Analyst for Urban District of —. 27.
- *Nickel** alloys; Determination of aluminium, magnesium or beryllium in —. R. C. Chirnside, L. A. Dauncey and P. M. C. Proffitt, 446.
- *Cyanometric** determination of cobalt and —. W. J. Agnew, 643.
- in foods and biological materials; Bibliography on —. 603.
- Titration of — with dimethylglyoxime with the aid of the dropping mercury electrode as indicator electrode. I. M. Kolthoff and A. Langer, 476.
- Nicotine**: Rapid method for determining —. A. Verda and E. Herzfeld, 234.
- Nicotinic Acid**: Chemical estimation of —. Inhibitory effect of cyanogen bromide upon the aniline side reactions. D. Melnick and H. Field, 661.
- in animal tissues, blood and certain food-stuffs; Estimation of —. E. Kodicek, 522.
- in biological materials estimated by means of photoelectric colorimetry. D. Melnick and H. Field, 571.
- in urine; Estimation of —. L. J. Harris and W. D. Raymond, 183.
- in urine; Quantitative estimation of —. E. Bandier, 182.
- Niger Seed Oil**: Component glycerides of —. N. L. Vidyarthi and M. V. Mallya, 419.
- Nitrate**: Function of — in curing of bacon and hams. 355.
- soil —; Variation in. 34.
- Nitric Acid**: New method for removing —. E. Müller and C. Burchard, 379.
- Nitrite**: Function of — in curing of bacon and hams. 355.
- in solution and in tissues; Manometric estimation of —. J. Brooks and J. Pace, 468.
- poisoning. 602.
- Nitrite—continued**
- Spot test for —. C. L. Wilson, 478.
- Nitro** compounds; Detection of ammonia, calcium and strontium with organic —. R. Fischer, 135.
- m-Nitrobenzamide** as reagent for identifying phenols. P. P. T. Sah and T. F. Woo, 124.
- o-Nitrobenzamide** as reagent for identifying phenols. P. P. T. Sah and W.-H. Yin, 373.
- p-Nitrobenzamide** as reagent for identifying amines. P. P. T. Sah, 374.
- p-Nitrobenzenesulphonic acid**: Detection of magnesium by means of —. J. P. Mehlig and K. R. Johnson, 314.
- Nitrogen** in coal and coke; Determination of —. 648.
- Nitro-β-Naphthol**: Non-precipitation of cobalt and palladium by —. C. Mahr and W. Prodingier, 187.
- o-Nitrophenol** in *p*-nitrophenol; Estimation of — by fluorescence analysis. W. Seaman, A. R. Norton and O. E. Sundberg, 581.
- p-Nitrophenyl Isocyanate** as reagent for identifying amines. P. P. T. Sah, 374.
- Nitrous Acid** method for determining amino-nitrogen; Behaviour of some uramido acids in —. A. G. Gornall and A. Hunter, 467.
- Nitrous Oxide** for whipped cream. 651.
- Njugo Bean**: J. M. Holm and B. W. Marloth, 608.
- Norfolk**: Appointment of W. F. Greeves as Deputy Public Analyst for County of —. 566.
- Northern Ireland**: Application of Food and Drugs Act, 1938, to —. 40.
- Norwegian Canning Industry**: Olive oil specifications for —. R. Mathiesen, 296.
- Significance of bacterial count in determining freshness of herring and brisling, the raw materials of kippered herrings and canned brisling. E. Aschehoug and R. Vesterhus, 417.
- Novocaine**: Detection of —. F. Biedebach and H. Wiegand, 173.
- Nuneaton**: Appointment of F. G. D. Chalmers as Public Analyst for Borough of —. 161.
- Nutrient** media; Silicic acid —. O. Hettche, 243.
- Nutrition**: Milk and —; Report of Committee on. 31.
- Nutshells**: Composition of certain —. M. Phillips and M. J. Goss, 608.

O

- Oak** wood; Hemicelluloses of English —. M. H. O'Dwyer, 475.
- "Oat Feed"**: Composition of —. 462.
- Obituary Notices**: Bloxam, Hugh Charles Loudon, 437.
- Burgess, William Thomas, 1.
- Edmed, Frank George, 79, 391.
- Gladding, Thomas Stantial, 391.
- Morgan, Sir Gilbert, 79, 324.
- Murphy, Albert John, 587.
- Pingstone, George Arthur, 198.
- Pope, Sir William Jackson, 258.
- Rawson, Christopher, 437.
- Shutt, Frank T., 79, 392.

Obituary Notices—continued

Thorpe, Jocelyn Field, 483.

White, John, 325.

Odour lead poisoning. 36.

Oestriol: New colour reaction for —. C. Bachman, 118.

Oestrogens: Photometric determination of —. I. Modified Kober reaction for determining total — in a mixture of oestrogenic steroids. II. New colour reaction for oestriol. C. Bachman, 118.

Oestrone in extracts of human urine; Colorimetric assay of weakly phenolic ketones, —, in. N. B. Talbot and Others, 609.

Odium lactis in yeast; Estimation of —. 62.

Oil(s): Analysis of commercial —. Report of American Chemical Society Committee, 508.

Application of Bellier and Blarez tests to some —. R. Marcille, 170.

Arylation of —. Synthesis of tolyl stearic acid and its esters. W. Kimura and J. Tsurugi, 186.

Behaviour of — with antimony trichloride. W. H. Dickhart, 421.

Chaulmoogra —; Analysis of. III. *Hydnocarpus wightiana* oil. H. I. Cole and H. T. Cardoso, 45.

drying-; Oxidation of —. VI. Properties of peroxide, ketol and oxido groupings, including those of some resins. R. S. Morrell and E. O. Phillips, 615.

Essential —. See **Essential Oils**.

Fatty —. See **Fatty Oils**.

from *Cerberum odallum* kernels; Constants of —. 37.

from fruit of *Ferula alliacea*. P. K. Bose and S. N. Dutt, 421.

from sapota seeds; Fatty acids and glycerides of —. N. L. Vidyarthi and V. Mallya, 171.

from seeds of *Canarium commune* L. A. Steger and J. van Loon, 230.

from seeds of *Veronia anthelmintica*; Occurrence of an isomer of ricinoleic acid in —. N. L. Vidyarthi and M. V. Mallya, 186.

Liver — of some terrestrial animals. M. Tsujimoto and H. Koyanagi, 231.

lubricating; Detection and determination of ethylene glycol in —. H. Lamprey, E. E. Sommer and A. D. Kiffer, 665.

motor-; Determination of sludge in used or oxidised —. C. G. Ludeman, 664.

"non-drying" —; Mixed unsaturated glycerides of. B. G. Gunde and T. P. Hilditch, 364.

of bitter almonds; Variations in —. 503.

*Peroxide accumulation rate of —. Relationship to Mackey test results and oxidation on textiles. W. Garner and M. Elsworth, 347.

Rice bran as raw material of —. T. Hidaka, 44.

Sulphated — and Allied Products: Their Chemistry and Analysis. (Review), D. Burton and G. F. Robertshaw, 318.

*textile —; Alkaline ageing test for. W. Garner, 219.

Oils—continued

tocopherol in —; Quantitative determination after saponification. A. Emmerie, 370.

Unsaponifiable matter in sulphated —. D. Burton and G. F. Robertshaw, 615.

Ointment(s): Bleach (anti-gas ointment No. 1) —. B.S.I. specification for. 463.

*Bleach —. D. D. Moir, 154.

Bleach —. G. H. Macmorran, 423.

Bleach —. (Legal Notes), 30.

Carbolic —. (Legal Notes), 222.

Oldbury: Appointment of H. E. Monk as Public Analyst for Borough of —. 27.

Oleander species; Poisonous principles of —. 37.

Oleic Acid: Chromatographic separation of palmitic and stearic acids from a mixture of palmitic, stearic and —. C. Manunta, 64.

Olive Oil: Application of Bellier test and Blarez test to —. R. Marcille, 170.

Behaviour of — with antimony trichloride. W. H. Dickhart, 421.

specifications for Norwegian canning industry. W. Mathiesen, 296.

Thermo-stability of —. 301.

***Olsson's Radiographic Technique** for assaying vitamin D₂. A. Z. Baker and M. D. Wright, 326.

Orange peels; Methylene blue reducing system of Palestine — investigated by the Thunberg system. L. Frankenthal, 662.

Oranges: Wastage of — in transport. 224.

Ors Analysis for Chemists and Colleges; Technical Methods of —. 11th Ed. (Review), A. J. Weinig and W. P. Schoder, 194.

concentrates; Assay for platinum metals in —. J. Seath and F. E. Beamish, 310.

Organic acids; Iodimetric determination of —. B. Singh and S. Singh, 186.

analysis. Abstracts: 64, 123, 186, 245, 306, 373, 430, 473, 527, 575, 614, 663.

Chemicals; B.P. Sub-Committee on —. Recommendations, 167.

Chemistry; Introduction to Practical — (Review), F. G. Mann and B. S. Saunders, 74.

compounds containing oxygen; Qualitative test for —. D. Davidson, 306.

compounds; Detection of elements in —. L. Rosenthaler, 307.

compounds; Determination of arsenic in —. D. T. Lewis and V. E. Davis, 531.

compounds; Identification of —. II. Piperidyl derivatives of aromatic halogenonitro compounds. M. K. Seikel, 530.

*compounds; Micro-tests for elements in —. II. Phosphorus, arsenic and antimony. C. L. Wilson, 405.

nitro compounds; Detection of ammonia, calcium and strontium with —. R. Fischer, 135.

substances; Volumetric determination of — by chromic oxidation: Nitro-chromic solutions. M. H. Cordebar, 123.

Organisms in yeast; Estimation of foreign —. K. E. Jensen, 62.

- Organs:** tocopherol in animal —; Estimation of. P. Karrer, W. Jaeger and H. Keller, 427.
- Orpington:** Appointment of F. W. F. Arnaud as Public Analyst for Borough of —, 27.
- Oxalate** formation in ascorbic acid solutions. A. E. Jurist and W. G. Christiansen, 184.
- Oxford:** Appointment of J. H. Weber as Deputy Public Analyst for County Borough of —, 566.
- Oxidase** measurements; Use of ascorbic acid as substrate in —. B. D. Egell and F. Gerhardt, 469.
- Oxycelluloses:** Microchemical determination of reducing power of — (micro-copper, microferricyanide and micro-iodine numbers). E. Geiger and G. Müller, 618.
- of the acidic type; Adsorption of substantive dyes by —. S. M. Neale and W. A. Stringfellow, 247.
- Oxygen:** organic compounds containing —; Qualitative test for. D. Davidson, 306.
- Separation of *para*-hydrogen from —. E. Bergmann, L. Farkas and L. Sandler, 244.
- Wet combustion method for determining carbon and — consumed. B. E. Christiansen and J. F. Facer, 64.
- Ozone:** Starch-iodide method for —. C. E. Thorp, 525.
- P**
- Paints:** Lead compounds in cellulose spray —, 292.
- Palestine** orange peels; Methylene blue reducing system of — investigated by the Thunberg system. L. Frankenthal, 662.
- Report of Government Analyst for — for 1938. G. W. Baker, 166.
- Palladium:** Non-precipitation of — by nitro- β -naphthol. C. Mahr and W. Prodingier, 187.
- *Palm Oil:** Carotene determined in —, 276.
- Composition of commercial —. V. Partial separation of — by crystallisation as an aid to determination of the component glycerides. T. P. Hilditch and L. Maddison, 517.
- Thermo-stability of —, 301.
- Palmitic Acid:** Chromatographic separation of — from a mixture of oleic, stearic and —. C. Manunta, 64.
- Pantocaine:** Identification of —. F. Biedebach and H. Wiegand, 173.
- Pantothenic Acid:** Assay method for —. D. Pennington, E. E. Snell and R. J. Williams, 661.
- Papain:** Activation of —. J. S. Fruton and M. Bergmann, 303.
- *Paper:** Sampling of printed —. J. H. Young, 596.
- Paper-Board:** bacterial content of —; Routine determination of. L. C. Cartwright and S. S. Epstein, 612.
- Paprika:** Quantitative estimation of coloured components in —. L. Cholnoky, 232.
- Paraffin** as "macassar" oil. 165.
- Paraffin Oil** on currants and raisins. 164.
- *Paraffins:** Iodine value of soft —. H. Brindle, 409.
- Pasteurisation** of cream by the "flash" process. 600.
- Pastures:** Cobalt deficiency in —, 33.
- Patiala (Punjab):** Report of Chief Chemist for 1939-40. K. L. Budhiraja, 652.
- Pea seed;** Distribution of manganese in — in relation to marsh spot. H. H. Glasscock and R. L. Wain, 372.
- Pears:** Arsenic in —, 229.
- Pectic** substances of plants. VI. E. W. Bennison and F. W. Norris, 67.
- Pectins:** Relation between jelly strength, viscosity and composition of various —. E. W. Bennison and F. W. Norris, 67.
- Pelletierine** of commerce. J. A. Goodson, 422.
- Penicillium carmino-Violaceum** Bourge: Colouring matters of —, and production of ergosterol by —. H. G. Hind, 470.
- Constitution of carviolin, a colouring matter of —. H. G. Hind, 470.
- Penicillium cycloptum.** Westling: ω -hydroxy-emodin: a metabolic product of —. W. K. Anslow, J. Breen and H. Raistrick, 470.
- Percaine** confused with procaine. 600.
- Perochlorate:** Identification with Zwicker's reagent. G. H. Wagenaar, 533.
- Perochloric Acid:** Reaction of gallium with —. L. S. Foster, 68.
- Periodate:** Determination of glycerol with —. L. Voris, G. Ellis and L. A. Maynard, 425.
- Periodic Acid** as means of determining lactic and pyruvic acids. R. Boisson, 473.
- Permanganate** acid ashing micro-method for iodine determinations. I. Values in blood of normal subjects. D. S. Riggs and E. B. Man, 579.
- *Peroxide** accumulation rate of oils—Relationship to Mackey test results and oxidation on textiles. W. Garner and M. Elsworth, 347.
- Peroxides:** organic —; Determination of. H. A. Liebhafsky and W. H. Sharkey, 474.
- Perspiration:** Human foot — and upper boot leather. A. Colin-Russ, 659.
- Persulphate:** Identification with Zwicker's reagent. G. H. Wagenaar, 533.
- Petri Dishes:** B.S.I. specification for —, 463.
- Petroleum** acids; Separation and characterisation of —. H. G. Schultze, B. Shive and H. L. Lochte, 529.
- Conversion of —. (Review), A. N. Sachanen, 671.
- pH:** Stability of bacteria in relation to —. J. G. Baumgartner and G. G. Knock, 371.
- Pharmaceutical Chemistry;** Practical —, 4th Ed. (Review), F. N. Appleyard and C. G. Lyons, 436.
- products; Micro-determination of ethyl alcohol in —. Determination of chromic index. A. Ionescu-Matiu, C. Popesco and O. Constantinesco, 135.
- Pharmacognosy:** Report of Committee on Pharmacy and —, 107.
- Pharmacopoeia:** See *British Pharmacopoeia*.

- Pharmacy:** Report of Committee on Pharmacognosy and —. 107.
- Phenarasin Chloride:** New colour reaction of —. J. Delga, 309.
- Phenol ointment.** H. Brindle, 608.
- Phenolic compounds;** Identification and determination of —. L. F. Levy, 430.
- *Phenols:** Functions and reactions of —, in relation to hair dyes. H. E. Cox, 393.
Identification of — with *o*-nitrobenzazide as reagent. P. P. T. Sah and W.-H. Yin, 373.
m-Nitrobenzazide as reagent for identifying —. P. P. T. Sah and T. F. Woo, 124.
- Phosgene:** Detection of — in industry. D.S.I.R. Leaflet No. 8. 290.
- Phosphatases:** Effect of zinc on alkaline —. E. Hove, C. A. Elvehjem and E. B. Hart, 572.
- *Phosphate:** Gravimetric determination of —. D. T. Lewis, 560.
in qualitative analysis; Detection and elimination of — by means of zirconium salts. F. K. Pittman, 669.
in soil; Determination of available —. 35.
separation in qualitative analysis. J. N. Friend, R. H. Vallance and H. J. G. Challis, 578.
Spectroscopic determination of —. T. Török, 383.
- *Phosphates:** quantitative and qualitative analysis of —; New group separation for. G. J. Austin, 335.
- Phosphoric Acid:** aluminium in presence of iron and —; Determination of. G. Balanescu and M. D. Motzoc, 188.
Removal of — from hydrogen peroxide. S. R. Dickman and R. H. Bray, 477.
- Phosphorus compounds in soil.** 104.
Estimation of —. R. J. L. Allen, 519.
Erratum, 663.
in coal and coke; Determination of —. 648.
- *in organic compounds;** Micro-tests or —. C. L. Wilson, 405.
- *in titanium steels;** Determination of —. A. T. Etheridge and D. G. Higgs, 496.
- Influence of — on lead in blood and bone.** A. E. Sobel and Others, 235.
- vitamin D in food substances containing —;** Estimation of. K. H. Coward and E. W. Kassner, 523.
- Photoelectric colorimetry as means of estimating nicotinic acid in biological material.** D. Melnick and H. Field, 571.
- Photographic silver-gelatin paper as a reagent in spot analysis.** G. Schwarz, 619.
- Photometer:** KWSZ — for the determination of hydrocyanic acid. J. T. Sullivan, 122.
- Phthalisation in presence of pyridine.** S. Sabetay, 125.
- α -Phylloquinone:** Blue alkali salts of —. P. Karrer, 60.
- Physical methods of analysis.** Abstracts: 73, 136, 191, 253, 382, 435, 534, 581, 670.
- Physico-Chemical Methods.** Vol. I. Measurement and Manipulations. Vol. II. Practical Measurements. (Review), 384.
- Physiological Chemistry; Essentials of —.** (Review), A. K. Anderson, 76.
- Phytic Acid and the rickets-producing action of cereals.** D. C. Harrison and E. Mellaby, 113.
*content of some poultry feeding stuffs. R. H. Common, 79.
- Picric Acid method for determining hydrocyanic acid.** J. T. Sullivan, 122.
- Picolonates:** Microscopy of —. R. Dunn, K. Inouye and P. L. Kirk, 252.
- Pig:** Body fats of the —. V. Component glycerides of perinephritic and outer back fats from the same animal. T. P. Hilditch and W. H. Pedelty, 569.
- Pigments:** Analysis of chrome green and similarly prepared —. J. H. Van der Meulen, 133.
- Pine seedlings disease and silver.** 292.
- Piperazine:** Determination of —. A. Castiglioni, 422.
- Piperidyl derivatives;** Identification of aromatic halogenonitro compounds as —. M. K. Seikel, 666.
derivatives of aromatic halogenonitro compounds; Identification of —. M. K. Seikel, 530.
- Plant extracts;** Tillmann reaction for vitamin C in —. C. Gatti and A. Knallinsky, 367.
families; Distribution of vitamin B₁ in some —. M. Pyke, 470.
foodstuffs; Combined ascorbic acid in —. J. C. Pal and B. C. Guha, 522.
insecticide materials from Empire sources. 574.
material; Determination of iodine in —. G. S. Fraps and J. F. Fudge, 305.
tissues; Quantitative analysis of — for lithium by the Ramage flame spectrographic method. N. L. Kent, 581.
- Plants:** Boron in — determined by means of quinalizarin. K. C. Berger and E. Truog, 123.
leguminous fish-poison —; Active principles of. IV. Isolation of malaccol from *Derris malaccensis*. S. H. Harper, 472.
Linolenic acid in leaves of land —. M. Tsujimoto, 664.
Pectic substances of —. VI. Relation between jelly strength, viscosity and composition of various pectins. E. W. Bennison and F. W. Norris, 67.
Water supply to —. 105.
- Plasmaquin:** Identification of — by a colour reaction. A. E. Tchitchibabine and Ch. Hoffman, 47.
- *Plating solutions;** Analysis of cadmium —. G. S. Smith, 87.
- Platinum metals in ore concentrates;** Assay for —. J. Seath and F. E. Beamish, 310.
- *Plums:** Composition of —. 549, 553.
Fruit gumming of Victoria —. 512.
ripening of — by acetylene; Stimulation of. 225.
Rôle of sorbitol in the carbon metabolism of Kelsey —. I. Donen, 178.
- Poisoning by datura.** 36.
by *Manihot utilisima* (jungle potato). 36.
by *Melia composita*. 36.
by nitrites. 602.
by oduvan leaf. 36.
by quinine hydrochloride. 652.

- Poisoning**—*continued*
 by sulphanilamide; Detection of ——. 226.
 cases in Ceylon. 652.
 Caustic soda ——. 459.
 Formic acid ——. 459.
 in India, 601, 602.
 Industrial ——. (British Encyclopaedia of Medical Practice, Vol. 12). (Review), D. Hunter, 139.
 with lead and arsenic; Industrial ——. 291.
- Poisonous** fumes from an arc light. 292.
 principle of zamia fruit. 292.
 substance in bamboo shoots. 37.
- Poison(s)**: African arrow ——. *Adenium somalense* Balf. fil. M. Hartmann and E. Schlitter, 428.
 Home Office Statutory Rules and Orders, 1940, Nos. 452 and 453. 412.
 Irritant — in well water. 459.
- Polyalcohols** in presence of each other; Quantitative determination of certain ——. N. Allen, H. Y. Charbonnier and R. M. Coleman, 575.
- Porcelain**: B.S.I. tests for laboratory ——. 516.
- Pork** in Hamburg steak. 227.
- Porous** bodies; Properties of ——. 598.
- Porphyrindin**: Determination of reducing groups with —, with special reference to egg albumin. E. Brand and B. Kassell, 425.
- Potassium**: Micro-determination of ——. A. Kaye, 533.
- *Potassium Ferricyanide** in sodium carbonate solution; Potentiometric estimation of Glucose with ——. H. T. S. Britton and L. Phillips, 149.
- Potassium Permanganate**: Toxicology of ——. P. Cheramy and A. Lemos, 120.
- Potassium Salts** precipitated by uranyl acetate reagents for sodium; Composition of ——. E. R. Caley and W. O. Baker, 131.
- Potassium Thiocyanate**: chlorides in —; Detection of. A. Slooff and D. Van Duyn, 250.
- Potato** carbohydrates; Metabolism of ——. 224.
- *Potentiometric** estimation of glucose with potassium ferricyanide in sodium carbonate solution. H. T. S. Britton and L. Phillips, 149.
- *Poultry** feeding stuffs; Phytic acid content of some ——. R. H. Common, 79.
- *Praseodymium**: Estimation of neodymium and — in solution from their absorption spectra. J. N. Friend and D. A. Hall, 144.
- Preservative** mixture for wood. 163.
 value of hops; Colorimetric determination of ——. A. A. D. Comrie, 516.
- Preservatives**: *n*-Butyl-*p*-hydroxybenzoate as ——. 291.
 for preparations containing gelatin. L. Gershenfeld and D. Perlstein, 62.
 in food; Public Health Amendment Regulations, 1940. 359.
- Pressure Cooker**: Effectiveness of heat penetration in the canning of meat in the home by ——. C. I. Nelson and D. Berrigan, 120.
- Printing Ink**: Examination by ultra-violet light. 38.
- Procaine** confused with percaine. 600.
- Producer-Gas** generator fuels; Use of certain South African woods as ——. P. E. Hall, 577.
- Prolactin**: International standards for ——. 40.
- *Prontosil**: Determination of ——. 211.
- Propylene Glycol** as vehicle for essences. 599.
- Proseptasine**: Determination of ——. 212.
- Protein** hydrolysates; Isolation of hydroxy-amino-acids from ——. R. L. M. Synge, 238.
- Proteins**: Glutamic acid of normal and malignant tissue ——. A. C. Chibnall and Others, 520.
 *on surface of metal plates; Detection of small amounts of ——. J. H. Young, 564.
 serum-; Basic amino acid content of ——. Influence of ingestion of arginine on composition of ——. R. J. Block, 303.
 serum-; Biuret reaction in estimating ——. H. W. Robinson and C. G. Hogden, 466.
- Proteolytic** activity; Polariscopic determination of ——. Q. Landis, 660.
- Public Analysts**: Notes from Reports of ——. *See Aberdeen, Birmingham, Cardiff, Kent, Salford, Stepney.*
 Society of ——. *See Society of Public Analysts.*
- Public Health** (Preservatives, etc., in Food) Amendment Regulations, 1940. 359.
- Pulp** fibres; Cross sections of ——. J. H. Graff, M. A. Schlosser and E. K. Nihlen, 670.
- Pyrethrin I**: Determination of ——. Seil colour reaction in the mercury reduction method. C. S. Sherman and R. Herzog, 373.
- Pyrethrum** from different sources; Analyses of ——. 574.
- Pyridine**: Phthalisation in presence of ——. S. Sabetay, 125.
- Pyrocatechol**: Activation of vanadium with ——. L. Szebelledy and M. Ajtai, 191.
- Pyruvic Acid**: Determination of — by means of periodic acid. R. Boisson, 473.
 in blood; Stabilisation and determination of ——. E. Bueding and H. Wortis, 426.

Q

- Qualitative** Analysis; Laboratory Manual of ——. (Review), J. H. Yoe, 254.
- Quantitative** Analysis; Calculations of ——. (Review), C. Engelder, 674.
 Chemical Analysis; Text-book of ——. 7th Ed. (Review), A. C. Cumming and S. A. Kay, 138.
 Inorganic Analysis; Text-book of ——. (Review), A. I. Vogel, 195.
 micro-determinations with the use of ordinary analytical balances. A. A. Benedetti-Pichler and R. A. Paulson, 69.
- Quartz** particles in felspar; Optical method for determining ——. G. H. McIntyre and M. Bozain, 535.
- Queensland**: Report of Government Analyst to — for 1938-39. F. E. Connah, 164.
- Quinaldine Iodobismuthite**: Determination of bismuth by precipitation as ——. J. R. Hayes and G. C. Chandlee, 129.
- Quinaldinic Acid** as a micro-reagent. IV. Determination of zinc in presence of copper, silver and mercury. P. R. Ráy and T. C. Sarkar, 435.

Quinaldinic Acid—continued

*as reagent for separating copper and cadmium. A. J. Lindsey and R. J. Shennan, 636.

Quinine and cinnamon tablets, 29.

in biological fluids and tissues; Colorimetric estimation of —. R. O. Prudhomme, 239.

in quinine iodobismuthate and its injectable preparations; Determinations of —. G. N. Thomas and G. P. Kopanaris, 49.

poisoning, 652.

Quinine Iodobismuthate: bismuth, iodine and quinine in —; Determination of. G. N. Thomas and G. P. Kopanaris, 49.**Quinoline:** Extraction of coal with —. R. Belcher and R. V. Wheeler, 614.**R****Rabbit Liver Oil:** 231.

Rabbits killed by hydrogen cyanide gas; Toxicity of flesh of —. 104.

Radix pereirae bravae: Origin of —. H. King, 518.**Ragwort:** Chemical control of —. 35.**Raisins:** Paraffin oil on —. 164.

Ramage flame spectrographic method; Quantitative analysis of plant tissues for lithium by —. N. L. Kent, 581.

Raman Effect and its Chemical Applications. (Review), J. H. Hibben and E. Teller, 192.**Rancidity of herring oil.** 224.

Rapeseed Oil: Brassicasterol, the characteristic sterol of —. E. Fernholz and H. E. Stavel, 572.

Rare Earths: Microchemistry of —. G. Beck, 382.***Raspberries:** Composition of —. 543, 547.

Rat virus enquiry report from Malaya. J. T. Paranjothy, 460.

Rayon: Estimation of viscose —. R. W. McKay, 432.

Reagent(s): cuprous iodide —; Action on alkaloïds: Precipitation and colour reactions. M. Péronnet and J. Guénin, 301.

for amino acids; Naphthalene-sulphonic acid as —. M. Bergmann and W. H. Stein, 237.

for identifying amines; 3·5-Dinitro-4-methylbenzazide as —. P. P. T. Sah, 125.

for identifying amines; *p*-Nitrobenzazide and *p*-nitrophenyl isocyanate as —. P. P. T. Sah, 374.

for identifying phenols; *m*-Nitrobenzazide as —. P. P. T. Sah and T. F. Woo, 124.

for identifying phenols; *o*-Nitrobenzazide as —. P. P. T. Sah and W.-H. Yin, 373.

*for separation of copper and cadmium; Quinaldinic acid as —. A. J. Lindsey and R. J. Shennan, 636.

for spot analysis; Photographic silver-gelatin paper as —. G. Schwarz, 619.

Grignard — for determining hydroxyl groups. W. Fuchs, N. H. Ishler and A. G. Sandhoff, 663.

Reagents—continued

Grignard — for micro-gravimetric determination of active hydrogen. R. N. Evans, J. E. Davenport and A. J. Revukas, 533.

Grignard — prepared from magnesium amalgams. E. G. Rochow, 245.

Modified Deniges — for determining methylpropene. A. Newton and E. J. Buckler, 473.

Quinaldinic acid as micro- —. P. R. Ray and T. C. Sarkar, 435.

Zwicker's — for identifying perchlorate, persulphate and other inorganic acid radicles. Sensitive reaction for copper. G. H. Wagenaar, 533.

***Redcurrants:** Composition of —. 544, 547.

Reducing Groups: Determination of — with porphyrindin, with special reference to egg albumin. E. Brand and B. Kassell, 425.

Refractive Index of non-opaque minerals; Immersion liquids for determining —. E. P. Kaiser and W. Parrish, 137.

Refuse: Use of town — as manure. 104, 221.

Reptiles: Vitamin A and carotenoids in the liver of —. H. B. Jensen and T. K. With, 179.

Resins: Properties of peroxide, ketol and oxido groupings, including those of some —. R. S. Morrell and E. O. Phillips, 615.

synthetic — containing maleic acid; Analysis of —. E. Sadolin, 128.

Rhenium in molybdenum; Determination of —. C. F. Hiskey and V. W. Meloche, 669.

Riboflavin content of foodstuffs; Fluorometric method for determining —. A. Z. Hodson and L. C. Norris, 114.

Estimation of —. J. O. Irwin and Others, 522.

Rice: Antineuritic value of parboiled —. I. A. Simpson, 461.

bran as a raw material of oil. T. Hidaka, 44.

Differentiation of hand-pounded from milled —. 602.

factor; Identification of —. H. J. Almquist and Others, 573.

Ricinoleic Acid: Occurrence of an isomer of — in the fatty oil from the seeds of *Vernonia anthelmintica*. N. L. Vidyarthi and M. V. Mallya, 186.

Rickets-producing action of cereals; Phytic acid and —. D. C. Harrison and E. Mellanby, 113.

Ripening of plums stimulated by acetylene. 225.

Rocks: alkalis in —; Modification of J. L. Smith method for extracting. R. E. Stevens, 616.

Cobalt in New Zealand —. 34

Root-forming Substances used for propagation purposes. M. A. H. Tincker and C. H. Unwin, 119.

Rose Hips: Ascorbic acid content of —. W. Goldberg and E. O'F. Walsh, 304.

Rosin size; Analysis of —. E. A. Georgi, 614.

Rotenone in derris root; Determination of —. T. M. Meijer and D. R. Koolhaas, 526.

Rothamsted Experimental Station: Report for 1938. 104.

- Royal Agricultural Society of England:** Report of Consulting Chemist for 1939. E. Voelcker, 462.
- Rubbing** of dyed materials; Machine for testing fastness to —, D. A. Derrett-Smith and H. B. Bradley, 534.
- Rugby:** Appointment of W. T. Rigby as Public Analyst, and F. G. D. Chamlers as Deputy Public Analyst for Borough of —, 27.
- Ruthenium:** Determination of — with thionalide. W. J. Rogers, F. E. Beamish and D. S. Russell, 669.

S

- Saccharated** iron tablets. 565.
- Saccharin** in foodstuffs; Detection of —, W. F. Reindollar, 569.
- Salford:** Report of City Analyst for 1939. G. H. Walker, 503.
- Salicylaldehyde** derivatives; Qualitative reactions of — with inorganic ions. J. F. Flagg and N. H. Furman, 688.
- in the colorimetric determination of iron. D. E. Howe and M. G. Mellon, 578.
- Salmonella** enquiry. 225.
- Salts:** Blue alkali — of α -phyloquinone (vitamin K₁) and similar compounds. P. Karrer, 60.
- Neutral — and soils. 166.
- of complex cations applied to the microscopic detection of anions. Mono-aquopentamminocobaltic chloride (roseocobaltic chloride). W. A. Hynes and L. K. Yanowski, 70; 1,2-Chloro-quotetramminocobaltic chloride. L. V. Yanowski and W. A. Hynes, 190.
- present in soil and spray waters; Chemical effect on lead arsenate of —, J. M. Ginsburg, 613.
- Samsu:** Lead in —, 459.
- Sapota Seeds:** Fatty acids and glycerides of oil from —, N. L. Vidyarthi and V. Mallya, 171.
- Sausages** (Maximum Prices) Order, 1940. 257, 358; Amendment, 507.
- meat content of —; Determination of. 257.
- Soya-bean flour in — estimated by determining non-fermentable sugars. W. B. Hendrey, 109.
- Savonnier:** Manuel du —. (Review), A. Matagrín, 389.
- Scandium:** Determination of —, 382.
- Scarborough:** Appointment of R. Mallinder as Public Analyst for Borough of —, 411.
- Science:** Social Function of —. (Review), J. D. Bernal, 140.
- Scientific and Industrial Research, Dept. of.** Building Research Board Report for 1939. 598.
- Food Investigation Board Report for 1938. 223.
- Forest Products Research Board Report for 1938. 163.
- Fuel Research Report. Analysis of commercial grades of coal, Part II. 32.
- Methods for detecting toxic gases in industry. Leaflet No. 11, Aniline vapour, 162; Leaflet No. 7, Carbon monoxide, 223; Leaflet No. 8, Phosgene, 290; Leaflet No. 9, Arsine, 354.
- Scientific and Industrial Research, Dept. of—continued**
- Methods of analysis of coal and coke. Fuel Research Paper No. 44, 648.
- National Physical Laboratory Report for 1939. C. G. Darwin, 413.
- Water Pollution Research Board. Report for 1938-39, 101.
- Scotland:** Application of Food and Drugs Act, 1938, to —, 39.
- Seunthorpe:** Appointment of J. Evans as Borough Analyst for —, 161.
- Seed fats.** See **Fats**.
- Seil Colour Reaction** in the mercury reduction method of determining pyrethrin I. C. S. Sherman and R. Herzog, 373.
- *Selenious Acid:** Determination of thiourea by oxidation with —, A. E. A. Werner, 286.
- Selenium** compounds; Excretion of volatile — after administration of sodium selenite to white rats. J. Schultz and H. B. Lewis, 302.
- in Canadian wheat. J. G. Malloch, 430.
- Micro-titration of —, G. Wernimont and F. J. Hopkinson, 534.
- Minimum lethal dose of — as sodium selenite for horses, mules, cattle and swine. W. T. Miller and K. T. Williams, 613.
- Serum:** blood-; Factors preventing oxidation of ascorbic acid in —, E. M. Mystkowski and D. Lasocka, 59.
- gonadotrophic substance of pregnant mares' —; International standards for. 40.
- proteins; Basic amino acid content of —, Influence of ingestion of arginine on composition of —, R. J. Block, 303.
- proteins. Biuret reaction in estimating —, H. W. Robinson and C. G. Hogden, 466.
- Sesame Oil:** Thermo-stability of —, 301.
- *Sewage** effluents; Rapid stability test for —, D. Dickinson and W. M. Weir, 289.
- *liquors; Determination of chromates in —, D. Dickinson, 409.
- Treatment and disposal of —, 102.
- Sheep:** Ragwort feeding to —, 35.
- Shellfish:** Added water in —, 227.
- Silica** basins, crucibles and capsules; B.S.I. standard dimensions for —, 41.
- Colorimetric estimation of — and its use in metallurgy of aluminium. P. Urech, 131.
- in cryolite and materials containing fluorine; Determination of —, H. Spielhacsek, 315.
- in the hot-springs of Gastein; Determination of —, R. Bisanz and F. Kroupa, 189.
- Silicates:** alumina in —; Direct determination of. E. W. Koenig, 130.
- Silicic Acid** in biological substances; Photometric estimation of —, J. Bodnár and T. Török, 240.
- nutrient media. O. Hettche, 243.
- Silicon** in fluorspar and cryolite; Determination of —, T. Nakamura, 434.
- Silver:** Colorimetric determination of —, C. G. Makris and R. Menaché, 477.
- in catadynised vinegar and fruit cordials; Estimation of —, O. Noetzel, 364.
- in unhealthy pine seedlings. 292.

Silver—continued

- reductor; Determination of uranium and copper with the aid of ——. N. Birnbaum and S. M. Edmonds, 313.
- reductor; Reduction of molybdate in the ——. C. F. Hisekey, V. F. Springer and V. W. Meloche, 69.
- Spot tests for ——. E. A. Kocsis, 190.
- zinc in presence of ——. Quinaldinic acid as micro-reagent in determining. R. R. Ray and T. C. Sarkar, 435.
- Silver Foxes:** anti-grey hair vitamin for —; Studies on. G. Lunde and H. Kringstad, 366.
- Silver-Gelatin** photographic paper as reagent in spot analysis. G. Schwarz, 619.
- Size:** rosin —; Analysis of. E. A. Georgi, 614.
- Slag:** Utilisation of blast furnace ——. 598.
- Smith (J. Lawrence)** method for extracting alkalis in rocks; Modification of ——. R. E. Stevens, 616.
- Soap** Manufacturer's Manual. (Review), A. Matagrin, 389.
- Society of Public Analysts:** Annual Report of Council, 198.
- Proceedings: 1, 143, 197, 257, 323, 539, 587.
- North of England Section, 143, 202, 323.
- Scottish Section, 198, 202, 323.
- Soda Crystals:** Transport of ——. 165.
- Sodamint** tablets. 597.
- Sodium:** Colorimetric micro-method for estimating — with manganous uranyl acetate. E. Leva, 316.
- in biological fluids; Determination of ——. M. C. Darnell, Jr., and B. S. Walker, 478.
- in presence of other metals; Determination of ——. E. C. Elliott, 616.
- uranyl reagents for —; Composition of lithium and potassium salts precipitated by. E. R. Caley and W. O. Baker, 131.
- *Sodium Carbonate** solution; Potentiometric estimation of glucose with potassium ferricyanide in ——. H. T. S. Britton and L. Phillips, 149.
- Sodium Hydrosulphite:** Colorimetric estimation of ——. J. Lotichius, 668.
- Sodium Permanganate:** Toxicology of ——. P. Cheramy and A. Lemos, 120.
- Sodium Selenite:** Excretion of volatile selenium compounds after administration of — to white rats. J. Schultz and H. B. Lewis, 302.
- Minimum lethal dose of selenium as — for certain animals. W. T. Miller and K. T. Williams, 613.
- Soil(s):** Arsenic in ——. 34.
- available phosphate in —; Determination of. 35.
- base exchange capacity of —; Manganese method for determining. C. A. Bower and E. Truog, 614.
- Boron in — determined by means of quinalizarin. K. C. Berger and E. Truog, 123.
- Chemical effect of salts in — on lead arsenate. J. M. Ginsburg, 613.
- clods and crumbs; Determination of size distribution of —. E. W. Russell and R. V. Tamhane, 371.

Soil(s)—continued

- Cobalt deficiencies in ——. 105.
- Cobalt in New Zealand ——. 34.
- Colour of ——. 105.
- Electrodialysis of —. Recovery of manganese from —. R. C. Hoon and C. L. Dhawan, 526.
- in Waioatapu Valley, New Zealand; Arsenic in ——. R. E. R. Grimmer and I. G. McIntosh, 613.
- Iodine determination in ——. G. S. Fraps and J. F. Fudge, 305.
- Manganese deficiencies in ——. 104.
- minerals; X-ray analysis of ——. 105.
- Neutral salts and ——. 166.
- New cell for electrodialysis of ——. A. W. Marsden, 435.
- nitrate; Variation in ——. 34.
- Phosphorus compounds in ——. 104.
- treatment; Effect of — on vitamin B₁ content of wheat and barley. P. C. Leong, 59.
- *Use of electrodialysis in analysis of —. M. Puffeles, 601.
- *Soluseptasine:** Determination of ——. 212.
- Soot** deposit; Acidity of ——. 503.
- Sorbitol:** Role of — in the carbon metabolism of the Kelsey plum. I. Donen, 178.
- d-Sorbitol** in diabetic chocolate. 651.
- South Africa:** Fertiliser problems in vegetable production. E. J. Greenstein, 415.
- Report of Division of Chemical Services for 1939. J. P. van Zyl, 229.
- South African** coals; Application of rational analysis to some typical ——. G. K. Morrison, 376.
- Compost; its preparation and use. E. R. Orchard, 416.
- woods; Analyses of certain —, with special reference to their use as producer-gas generator fuels. P. E. Hall, 577.
- Sowans:** Composition of ——. 28.
- Soya-Bean** flour in sausage estimated by determining non-fermentable sugars. W. B. Hendrey, 109.
- husks. 462.
- Vitamin A activity and vitamin B₁ content of ——. J. O. Halverson and F. W. Sherwood, 470.
- Spectrographic** analysis with the microphotometer; Quantitative ——. D. M. Smith, 295.
- Spectroscopy:** Chemical —. (Review), W. R. Brode, 137.
- α-Spinasterol:** Isolation of — from alfalfa. E. Fernholz and M. L. Moore, 60.
- *Spirit Vinegar:** Analytical constants of —. C. H. Manley, 594.
- Spot Tests:** Detection of barium and sulphate by —. F. Feigl and W. Aufrecht, 251.
- for silver, lead, mercury, uranium and aluminium. E. A. Kocsis, 190.
- Photographic silver-gelatin paper as reagent for —. G. Schwarz, 619.
- Spray** paints; Lead compounds in cellulose ——. 292.
- residues; Toxicity to sheep of lead arsenate ——. J. L. St. John and Others, 575.

- Spray**—*continued*
test for determining relative atmospheric corrodibility of ferrous materials. T. Swinden and W. W. Stevenson, 670.
waters; Chemical effect on lead arsenate of certain salts present in —. J. M. Ginsburg, 613.
- Stains** in fabrics. 228.
- Stamps**: Re-used postage —; Examination of. 603.
Transference of revenue — from documents. 460.
- Standardisation** of testing methods for textiles. 653.
- Standards**: Carboxyhaemoglobin solution for use in haemoglobinometer. 414.
Electrical —. 414.
for brown and wholemeal bread. 649.
for cheese in New South Wales. 650.
for food; Report on —. 103.
for malted milk in New South Wales. 651.
for nephelometric determination of arsenic with Bougault's reagent; Permanent artificial —. J. Thuret, 129.
for prolactin and for the gonadotrophic substance of pregnant mares' serum. 40.
for trace elements in pastures. 34.
for tripe in New South Wales. 651.
for water for irrigation purposes. 292.
See also **British Standards Institution**.
- Starch** and Its Derivatives. (Review), J. A. Radley, 481.
banana —; Constitution of. E. G. E. Hawkins, J. K. N. Jones and G. T. Young, 464.
Characterisation of — from various Sources by means of electrophoretic fractionation. O. Dahl, 362.
in timber; Estimation of —. 164.
products; Simplified alkali-lability determination for —. T. J. Schoch and C. C. Jensen, 655.
- Starch-Iodide** method for ozone. C. E. Thorp, 525.
- Stearic Acid**: Chromatographic separation of — from a mixture of oleic, palmitic and —. 64.
Product of the dehydrogenation of —. O. St. A. K. Lang and H. Mayer, 241.
- Steel**: molybdenum in —; Determination of. C. Sterling and W. P. Spuhr, 311.
*phosphorus in titanium —; Determination of. A. T. Etheridge and D. G. Higgs, 496.
- Step-Photometer**: Zeiss — as means of estimating vitamin C. F. Bukatsch, 241.
- Stepney**: Report of the Borough Analyst for 1938. D. Henville, 29.
- Sterilisation** of water consumed by evacuated populations; Practical methods for —. R. D. de la Rivière, 429.
- Sterol** of rapeseed oil; Brassicasterol, the characteristic —. E. Fernholz and H. E. Stavely, 572.
- Sterol Acetate** test for ghee. 601.
- Stockton-on-Tees**: Appointment of C. J. H. Stock as Public Analyst for Borough of —. 411.
- Storage**: Gas — of fruit and vegetables. 224.
- *Strawberries**: Composition of —. 543, 546.
- Streptococcus mastitis** diagnosed by cultural methods. S. J. Edwards, 62.
- Stretford**: Appointment of G. H. Walker as Borough Analyst for —. 161.
- Strontium**: Detection of — with organic nitro compounds. R. Fischer, 135.
- *Succinic Acid** in beer. R. L. Andrew and L. G. Neubauer, 455.
Micro-determination of —. G. J. Goepfert, 580.
- Sucrase** activity in the barley plant. H. K. Archbold, 520.
- Suffolk, East and West**: Appointment of W. F. Greeves as Deputy Agricultural Analyst for County of —. 411, 566.
- Sugar(s)**: Blood —. *See* **Blood**.
Determination of non-fermentable — for estimating soya-bean flour in sausage. W. B. Hendrey, 109.
for chemical purposes; Permits to purchase —. 161.
in brewery effluents. 356.
in flour; Determination of —. W. Iwanowski and G. Grabouska, 168.
in urine; Relation of cuprous creatinine to tests for —. M. Samson, 55.
- Sulphanilamide**: Action of —. 428.
*group of drugs; Determination of members of —. G. V. James, 206.
poisoning; Detection of —. 226.
- Sulphate**: Detection of — by means of spot reactions. F. Feigl and W. Aufricht, 251.
Estimation of small amounts of sulphur as — in biological fluids. A. D. Marenzi and R. F. Banfi, 366.
in water; Volumetric determination of —. J. Courtois and P. Bonjean, 121.
Interference of sulphite with the determination of — by the tetrahydroxyquinone method. H. L. Kahler, 434.
Volumetric determination of —. A. Krüger, 434.
Volumetric determination of small quantities of — with barium rhodizonate as indicator. C. C. Miller, 477.
- *Sulphate of Alumina**: free acid in —; New method of determining. J. R. Simmons, 456.
- Sulphide** in Harrogate mineral water; Presence of — and its oxidation by air. A. Woodmansey, 430.
- Sulphide Sulphur** in presence of other sulphur compounds; Determination of —. J. B. Lewis, 669.
- Sulphides** in depolaritors; Determination of —. E. M. Hoshall, 579.
- Sulphite**: Interference of — with the determination of sulphate by the tetrahydroxyquinone method. H. L. Kahler, 434.
- Sulphur**: Colour test for elemental —. H. Sommer, 532.
compounds; Determination of sulphide sulphur in presence of other —. J. B. Lewis, 669.
compounds in vegetables; Estimation of —. W. Diemair and J. Koch, 418.
content of foods. M. Masters and R. A. McCance, 42.

Sulphur—continued

- in biological fluids; Estimation of small amounts of — as sulphates. A. D. Marenzi and R. F. Banfi, 366.
- in canned fruits; Behaviour of traces of —, 512.
- in coal and coke; Determination of —, 649.
- micro-determination of —; New method for washing and isolation of barium sulphate in. R. Grangaud, 316.
- *Separation of molybdenum from —, D. A. Lambie and W. R. Schoeller, 281.

Sulphur Dioxide: Effect of — on vegetation. G. S. Whitby, 51.

in beer. 599.

in mince. 28.

Summer Crook-Neck Squash: Ascorbic acid oxidase from —, P. L. Lovett-Janison and J. M. Nelson, 572.

***Supron:** Determination of —, 213.

Surbiton: Appointment of E. Hinks as Public Analyst and D. D. Moir as Deputy Public Analyst for Borough of —, 27.

Surgical Dressings: 228.

Swindon: Appointment of R. H. Ellis as Borough Analyst for —, 161.

Swinton and Pendlebury: Appointment of T. R. Hodgson as Public Analyst for Borough of —, 566.

T

Tablets: ascorbic acid content of —; Determination of. E. C. M. J. Hollman, 365.

Tan liquors; Use of cheap filter paper for analysing —, H. B. Merrill and G. C. Mueller, 476.

Tannic Acid: Tests for —, D. B. Dott, 233, 424.

Tannin materials; Estimation of lignin in —, J. G. Shrikhande, 531.

solutions; Determination of colour of vegetable —, 568.

Tea — and its fermentation products. C. J. Harrison and E. A. H. Roberts, 46.

Tannins: Precipitation of natural — with calcium hydroxide. V. Nemec, 246.

Tans: catechin- — in gambir; Determination of. C. J. van Hulssen and D. R. Koolhaas, 127.

Taraktogenos kurzii Oil: Analysis of —, H. I. Cole and H. T. Cardoso, 231.

Tattoo Marks: Identification of —, 602.

Tea manufacture; Fermentation process in —, IV. Tea tannin and its fermentation products. C. J. Harrison and E. A. H. Roberts, 46; V. Cytochrome oxidase and its probable rôle. VI. Effect of dilution on rate and extent of oxidations in fermenting tea leaf suspensions. E. A. H. Roberts, 521.

Tellurium in tin-rich alloys; Determination of — by volatilisation. W. T. Pell-Walpole, 434.

Temperature scale; International —, 413.

Tetrahydroxyquinone method for determining sulphate; Interference of sulphite with —, H. L. Kahler, 434.

Textile materials; Definitions of atmospheric humidity and moisture content and regain of —, 653.

materials; Determination of relative humidity. 654.

materials; Standard method for testing fastness to light of coloured —, L. A. Lantz, 582.

*scouring apparatus for the laboratory. W. Garner, 563.

***Textile Oils:** Alkaline ageing test for —, W. Garner, 219.

Textiles: Standardisation of testing methods for —, 653, 654.

Thermometers: General purpose laboratory —, B.S.I. specification for. 463.

Thiochrome method for estimating aneurin. R. G. Booth, 610.

Thiocyanate(s): Detection of —, D. Hart and R. Meyrowitz, 532.

in presence of chlorides and cyanides; Determination of —, A. Slooff and D. Van Duyn, 250.

Thiocyanogen absorption of triolein and tri-linolin. D. H. Wheeler, R. W. Riemen-schneider and C. E. Sando, 307.

Thiocyanogen Value of linolenic acid. J. P. Kass and Others, 528.

*Use of — in determining linoleic and linolenic acids and their glycerides. T. P. Hilditch and K. S. Murti, 437.

Thioglycolates in depilatories; Determination of —, E. M. Hoshall, 667.

Thionalide: Determination of ruthenium with —, W. J. Rogers, F. E. Beamish and D. S. Russell, 669.

***Thiourea:** Determination of — by oxidation with selenious acid. A. E. A. Werner, 286.

Thorium: Determination of —, 382.

Thorium Nitrate: Amperometric titration of fluoride with —, A. Langer, 669.

micro-titration of fluorine in aqueous and alcoholic systems. J. W. Hammond and W. H. MacIntire, 618.

Threonine: Micro-estimation of —, R. J. Block and D. Bolling, 72.

Thurrock: Appointment of A. W. Stewart as Public Analyst for Urban District of —, 411.

Thyroid and its preparations; Determination of iodine in — by cerate oxidimetry. W. W. Hilty and D. T. Wilson, 174.

Tillmann Reaction for vitamin C in plant extracts. C. Gatti and A. Knallinsky, 369.

Timber: Absorption of moisture by —, 163.

Fire resistance of —, 163.

Identification of —, 163.

starch in —; Estimation of. 164.

Tin coatings; Determination of lead in —, P. Foerster, 476.

in canned fish. 28.

*in foodstuffs determined by means of dithiol. R. de Giacomi, 216.

*Quantitative electro-deposition of — from chloride solutions. F. G. Kny-Jones, A. J. Lindsey and A. C. Penney, 498.

*Separation of molybdenum from —, D. A. Lambie and W. R. Schoeller, 281.

- Tin**—*continued*
 Tellurium in alloys rich in — determined by volatilisation. W. T. Pell-Walpole, 434.
- *Tin-Antimony Oxides:** Antimony —. A. G. Dunbar-Poole, 453.
- Tincture of Iodine:** Colourless —. 565.
- Tissue(s):** Ashing of soft — preliminary to determination of cations. M. V. Buell, 53.
 Combined ascorbic acid of animal —. P. Holtz, 367.
 fumurate and malate in animal —; Determination of. H. A. Krebs, D. H. Smyth and E. A. Evans, 571.
 nicotinic acid in animal —; Estimation of. E. Kodicek, 522.
 nitrite in —; Manometric estimation of. J. Brooks and J. Pace, 468.
 proteins; Glutamic acid of normal and malignant —. A. C. Chibnall and Others, 520.
 quinine in —; Colorimetric estimation of. R. O. Prudhomme, 239.
 Respiration of animal — after freezing in liquid air. F. Lynen, 425.
- Titanium** solutions; Stability of peroxidised —. G. H. Ayres and E. M. Vienneau, 379.
 *steels; Determination of phosphorus in —. A. T. Etheridge and D. G. Higgs, 496.
- *To bacco** ash; Spectrographic analysis of —. F. G. H. Tate and H. K. Whalley, 587.
- α -Tocopherol:** Oxidation products of —. L. I. Smith, W. B. Irwin and H. E. Ungnade, 115.
- Tocopherols:** Colorimetric determination of —. IV. Quantitative determination of — in oils after saponification. A. Emmerie, 370.
 in animal organs; Estimation of —. P. Karrer, W. Jaeger and H. Keller, 427.
 Red oxidation products of the —. W. John and W. Emte, 242.
- Toilet** preparations; Ammonia in —. 165.
- Tollens' Reaction:** Quantitative estimation of glycuronic acid and its conjugated compounds by means of —. W. Mozolowski, 519.
- Tolyl Stearic Acid** and its esters; Synthesis of —. W. Kimura and J. Tsurugi, 186.
- Tomato** products; Mould-content of —. 599.
 products; Rapid method for determining chlorides in —. L. M. Beacham, 109.
 pulp; Copper and zinc in —. 28.
 *Pulp; Copper in —. C. A. Mitchell, 220.
 pulp; Copper in concentrated —. 104.
 pulp; Detection of artificial dyes in —. H. Thaler and K. E. Schulte, 300.
 purée; Copper in —. 28.
- Tomatoes:** Copper in —. 28; W. J. Shannon and D. T. English, 605.
- Tooth** structure. 413.
- Tormentilla** *potentilla* flowers; Colouring matters of —. L. Schmid and A. Polacek-Witek, 309.
- Torquay:** Appointment of T. Tickle as Borough Analyst and C. V. Reynolds as Deputy Borough Analyst for —. 161.
- Total Solids:** Fat, — and Moisture. (Review). R. D. Mason, 75.
- Toxic** gases in industry; Detection of —.
 Aniline vapour. D.S.I.R. Leaflet No. 11. 162; Carbon monoxide. Leaflet No. 7. 223. Phosgene. Leaflet No. 8, 290; Arsine. Leaflet No. 9, 354.
- Toxicity** of coal tar naphtha distillates. H. Taylor, 121.
 of flesh of rabbits killed by hydrogen cyanide gas. 104.
 to sheep of lead arsenate and lead arsenate spray residues. J. L. St. John and Others, 575.
- Toxicological** alkaloids; Chemical microscopy of some —. W. F. Whitmore and C. A. Wood, 70.
 analysis. Abstracts: 63, 120, 243, 427, 470.
 detection of cocaine. A. Brünig, 427.
- Toxicology** of manganese. A. C. Lemos, 63.
 of sodium and potassium permanganates. P. Ceramy and A. Lemos, 120.
 Part 1 of British Encyclopaedia of Medical Practice, Vol. 12. (Review), G. Roche Lynch and D. M. Pryce, 139.
- Triethanolamine** in the colorimetric determination of copper. J. H. Yoe and C. J. Barton, 578.
- Trilinolin:** Properties, preparation and thiocyanogen absorption of —. D. H. Wheeler, R. W. Riemenschneider and C. E. Sando, 307.
- Trimethylene Glycol:** Formation of — from glycerol by *Aerobacter*. M. N. Nickelson and C. H. Werkman, 613.
- Triolein:** Preparation, properties and thiocyanogen absorption of —. D. H. Wheeler, R. W. Riemenschneider and C. E. Sando, 307.
 Thermo-stability of —. 301.
- Tripe:** Standard for — in New South Wales. 651.
- Tristearin** in lard; Detection of —. 508.
- Tuberculosis:** Contraphthisine; a reputed cure for —. 48.
- Tung Oil:** Reaction of Wijs' solution with —. S. W. Wan and D. B. Hu, 64.
- *Tungsten:** detection of —; New test for. J. H. Hamence, 152.
 in biological materials; Determination of —. J. C. Aull and F. W. Kinard, 660.
 Volumetric determination of —. M. L. Holt and A. G. Gray, 312.
- Turbidities:** Oxidation — in beer and their causes. E. Singruen, 569.
- Turkish** buffalo; Milk fat from —. A. Heiduschka and F. Cicekdagi, 297.
- L-Tyrosine** in alcoholic extract of egg yolk; Presence of —. L. Bracaloni, 298.

- Unsaponifiable matter**; Determination of hydrocarbons in —. J. Grossfeld, 375.
- Uramido Acids**; Behaviour of some — in the nitrous acid method for determining amino nitrogen. A. G. Gornall and A. Hunter, 467.
- Uranium** determination of — with the aid of silver reductor. N. Birnbaum and S. M. Edmunds, 313.
- Isotopes of —. 568.
- Micro-gravimetric separation of zinc and —. E. Kroupa, 435.
- Spot tests for —. E. A. Kocsis, 190.
- Uranyl Acetate** reagents for sodium; Composition of lithium and potassium salts precipitated by —. E. R. Caley and W. O. Baker, 131.
- *Uranyl Ammonium Vanadate**: 580.
- Urea**: B.P. Sub-Committee recommendation. 167.
- *in urine stains on carpets; Detection of —. G. N. Gee, 25.
- Urine**: adermin in —; Estimation of. J. V. Scudi, H. F. Koomes and J. C. Keresztesy, 304.
- aneurin in —; Chemical estimation, stability and form of. D. Melnick and H. Field, Jr., 56.
- arsenic in —; Colorimetric semi-microdetermination of. J. V. Harispe, 54.
- chloralose in —; Detection of. M. P. Cheramy, 471.
- distillates; Determination of alcohol in presence of acetone in —. 600.
- equol of mare's —; Colour reaction for detecting. W. Discherl, 368.
- Examination. (British Encyclopaedia of Medical Practice, Vol. 12). (Review). J. D. Robertson, 139.
- Fluorescence analysis of human —. W. Koschara, S. von der Seipen and P. A. Aldred, 242.
- histidine in —; Detection and estimation of. E. Racker, 467.
- lead in —; Determination of. 291.
- nicotinic acid in —; Estimation of. E. Bandier, 182; L. J. Harris and W. D. Raymond, 183.
- *Recovery of acetyl-sulphanilamidopyridine from —. 211.
- *stains on carpets; Detection of urea in —. G. N. Gee, 25.
- sugar in —; Relation of cuprous creatinine to tests for. M. Samson, 55.
- sulphur in —; Estimation as sulphates. 367.
- Urothione: a yellow sulphur-containing pigment from —. W. Koschara, 367.
- Uronic Acids**: Micro-estimation of —. E. M. Kapp, 580.
- Urothione**: a yellow sulphur-containing pigment from human urine. W. Koschara, 367.
- Vanillin** in vanilla extracts; Determination of —. H. J. Lynch and N. Deahl, 570.
- Vegetable** production in S. Africa; Fertiliser problems in —. E. J. Greenstein, 415.
- Vegetables**: Antiscorbatic values of —. M. Olliver, 662.
- Arsenic and lead limits for fresh —. 291.
- Canned —. *See Canned*.
- Colon bacilli in —. 227.
- Enzyme activity in frozen —: Asparagus. M. A. Joslyn and C. L. Bedford, 468.
- Gas storage of —. 224.
- sulphur compounds in —; Estimation of. W. Diemair and J. Koch, 418.
- Vegetation**: Effect of sulphur dioxide on —. G. S. Whitby, 51.
- Veratrum viride** powders; Evaluation of activity of — by the *Daphnia* method. I. Cohen, 184.
- Vernonia anthelmintica** seeds; Occurrence of an isomer of ricinoleic acid in the fatty oil from —. N. L. Vidyarthi and M. V. Mallya, 186.
- Vinegar** "essence." 565.
- silver in catadynised —; Estimation of. O. Noetzel, 364.
- *spirit —; Analytical constants of. C. H. Manley, 594.
- Virus** diseases. 105.
- for rats; Experiments on various types of —. J. T. Paranjothy, 460.
- Viscera**: Extraction of benzedrine from —. 5.
- Viscose** rayon; Estimation of —. R. W. McKay, 432.
- Visibility** curve; International —. 414.
- Vitamin A** activity of soya beans and cowpeas. J. O. Halverson and F. W. Sherwood, 470.
- content of cheese. A. W. Davies and T. Moore, 58.
- content of fish oils. 225.
- content of "light white" casein. M. K. Maitra and T. Moore, 116.
- content of visceral organs of the Geelbek or Cape salmon; Seasonal variations in —. C. J. Molteno and W. S. Rapson, 59.
- content of yolk of hens' eggs. B. Sjollem and W. F. Donath, 521.
- destruction in fish-liver oils. E. J. Simons, L. O. Buxton and H. B. Colman, 469.
- Estimation of — with the photoelectric colorimeter. C. J. Koehn and W. C. Sherman, 303.
- in the liver of mammals, birds, reptiles and man. H. B. Jensen and T. K. With, 179.
- *Spectrophotometric assay of —, with special reference to margarine. J. R. Edisbury, 484.
- Vitamin B₁**, content of soya beans and cowpeas. J. O. Halverson and F. W. Sherwood, 470.
- content of wheat and barley; Effect of soil treatment on —. P. C. Leong, 59.
- Distribution of — in some plant families. M. Pyke, 470.
- in cerebrospinal fluid; Estimation of —. H. M. Sinclair, 181.
- Vitamin B₂**, in urine; Estimation of —. J. V. Scudi, H. F. Koones and J. C. Keresztesy, 304.

- Vitamin C** content of honey. E. Becker and R. F. Kardos, 470.
 content of West Australian fruits. 291.
 Estimation of — by means of the Zeiss step-photometer. F. Bukatsch, 241.
 in foodstuffs; Estimation of —. P. N. Sen-Gupta and B. C. Guha, 523.
 in plant extracts; Tillmann reaction for —. C. Gatti and A. Knallinsky, 367.
- Vitamin D** in food substances containing phosphorus; Estimation of —. K. H. Coward and E. W. Kassner, 523.
 *in margarine; Estimation of —. N. T. Gridgeman, H. Lees and H. Wilkinson, 493.
 Influence of — on lead in blood and bone. A. E. Sobel and Others, 235.
- ***Vitamin D₂**: Biological assay of —. Methods in use with particular reference to Olsson's radiographic technique. A. Z. Baker and M. D. Wright, 326.
- Vitamin E** chemistry: Oxidation products of α -tocopherol and related 6-hydroxychromans. L. I. Smith, W. B. Irwin and H. E. Ungnade, 115.
 Colorimetric determination of —. IV. Quantitative determination of — in oils after saponification. A. Emmerie, 370.
 Oxidation of —. C. Golumbic and H. A. Mattill, 573.
 (Review). Food Group of Society of Chemical Industry, 77.
- Vitamin H**: Attempts to isolate the curative factor (—) for egg-white injury. P. György, R. Kuhn and E. Lederer, 117.
 Curative factor (—) for egg white injury with particular reference to its presence in different foodstuffs and in yeast. P. György, 117.
 Physicochemical properties of the factor (—) curative of egg white injury. T. W. Birch and P. György, 117.
- Vitamin K** activity of 4-amino-2-methyl-1-naphthol and 4-amino-3-methyl-1-naphthol. A. D. Emmett, O. Kamm and E. A. Sharp, 305.
- Vitamin K₁**: Blue alkali salts of —. P. Karrer, 60.
- Vitamin K₂**: Constitution of —. S. B. Binkley and Others, 524.
- Vitamin P**: H. Scarborough, 61.
- Vitamin(s)**: anti-grey hair — for silver foxes; Studies on. G. Lunde and H. Kringstad, 366.
 -free diets for animal experiments. A. I. Bacharach, 573.
 Part 3 of British Encyclopaedia of Medical Practice, Vol. 12. (Review), L. J. Harris, 139.
- Wakefield**: Appointment of F. W. Jaffé as Deputy Public Analyst for County Borough of —. 566.
- Walkiewicz's Method** for detecting decomposition of meat. H. Keller and H. Möller, 418.
- Wanstead and Woodford**: Appointment of B. Dyer and G. Taylor as Joint Public Analysts for Borough of —. 161.
- Warwickshire**: Appointment of F. G. D. Chalmers as Agricultural Analyst for County Council of —. 411.
- Water(s)**: Added — in shellfish. 227.
 analysis. Abstracts: 121, 245, 429.
 Arsenic in —. 34.
 Bacteria in chlorinated —. M. Levine, P. Carpenter and J. M. Coblentz, 370.
 Bacteriology of fresh —. 102.
 boiler — treatment; Application of hydrogen ion concentration to. G. W. Bond, 429.
 consumed by evacuated populations; Practical methods for sterilised —. R. D. de la Rivière, 429.
 distillation method for —; Effect of glycerin on. R. B. Trusler, 665.
 *drinking-; Estimation of lead in —. C. H. Manley, 403.
 drinking; Lead in —. 102.
 Fischer volumetric method for determining —. E. G. Almy, W. C. Griffin and S. C. Wilcox, 616.
 in ether; Determination of —. R. Gaspart and G. Serrure, 73.
 in Waiotapu Valley, New Zealand; Arsenic in —. R. E. R. Grimmett and I. G. McIntosh, 613.
 Iodine determination in —. G. S. Fraps and J. F. Fudge, 305.
 Mineral —. See **Mineral Water**.
 softening; Base exchange process of —. 101.
 Substance; Properties of Ordinary —. (Review), N. E. Dorsey, 621.
 sulphate in —; Volumetric determination of. J. Courtois and P. Bonjean, 121.
 supply to plants. 105.
 well —; Irritant poison in. 459.
- Water Pollution Research Board**: Report for 1938-39. 101.
- Wax**: Bamboo leaf —. M. Tsujimoto, 187.
- Wells** air centrifuge; Efficiency of — as determined by air-washing technique. K. MacDonald, 470.
- Western Australia**: Report of Chemical Branch, Mines Dept., for 1938. E. S. Simpson, 291.
- Westmorland**: Appointment of W. H. Roberts as Public Analyst for County of — within boundaries of Borough of Kendal. 411, 456.
- Wheat**: aneurin content of —; Thiochrome method for estimating. R. G. Booth, 610.
 Selenium in Canadian —. J. G. Malloch, 430.
 vitamin B₁ content of —; Effect of soil treatment on. P. C. Leong, 59.
- Widnes**: Appointment of J. R. Stubbs as Borough Analyst for —. 161.
- Wijs' Solution**: Reaction with tung oil. S. W. Wan and D. B. Hu, 64.
- Wimbledon**: Appointment of E. Hinks as Public Analyst and D. D. Moir as Deputy Public Analyst for Borough of —. 161.
- Windsor (New) Borough**: Appointment of J. H. Weber as Deputy Public Analyst for —. 161.

- Wine:** Arsenical insecticides and ——. 166.
 Natural ageing of ——. E. K. Nelson and D. H. Wheeler, 43.
- Wood:** Analyses of certain South African types of — with special reference to their use as producer-gas generator fuels. P. E. Hall, 577.
 -boring insects; Detection of — by X-rays. 164.
 -destroying fungi; Physiological studies of —. 164.
 fibres; Species identification of —. 577.
 insects in —; Destruction by fumigation. 164.
 moisture content of —; Effect of changes of. 163.
 of the English oak; Hemicelluloses of —. M. H. O'Dwyer, 475.
 Preservative mixture for —. 163.
 Species identification of —. 577.
 treated with fluoride preservative; Determination of fluorine in —. B. Ikert, 190.
- Woollen cloths;** Microscopical investigation of true —. Determination of additions of casein wool. J. Straub and G. J. Van Amerongen, 582.
- Worthing:** Appointment of S. A. Woodhead and R. F. Wright as Public Analysts for Borough of —. 27.
- Writing on a document;** Alterations to —. 603.

X

- Xanthophyll** content of yolk of hens' eggs. B. Sjollesma and W. F. Donath, 521.
- X-ray** analysis of soil minerals. 105.
- Xylose:** Blood sugar levels in rats receiving galactose and —. W. J. Darby and P. L. Day, 520.

Y

- Yeast** extracts; Ultra-violet absorption of —. J. Schormüller, 297.
 foreign organisms in —; Estimation of. K. E. Jensen, 62.
 in brewery effluents. 356.
 Manufacture of Compressed — (Review), F. G. Walter, 586.
Oidium lactis in —; Estimation of, 62.
 Presence of curative factor (vitamin H) for egg white injury in —. P. György, 117.
- *Yellow Corn:** Carotene determined in —. 277.
- Yorkshire, West Riding:** Appointment of E. Voelcker as Agricultural Analyst for County Council of —. 411.

Z

- Zamia Fruit:** Poisonous principle of —. 292.
- Zeiss** step-photometer as means of estimating vitamin C. F. Bukatsch, 241.
- Zinc:** Effect of — on alkaline phosphatases. E. Hove, C. A. Elvehjem and E. B. Hart, 572.
 in canned fish, 28.
 in canned vegetables. 651.
 in presence of copper, silver and mercury; Quinaldinic acid as microreagent in determining —. P. R. Ray and T. C. Sarkar, 435.
 in tomato pulp. 28.
 Micro-gravimetric separation of uranium and —. E. Kroupa, 435.
 Micro-determination of — by the hydroxy-quinoline method. IV. Separation from manganese, iron, bismuth, mercury, arsenic and antimony. C. Cimerman and P. Wenger, 381.
 Separation of cadmium from — by precipitation with aluminium. F. E. Townsend and G. N. Cade, 310.
- Zirconium Salts** as means for the detection and elimination of phosphate in qualitative analysis. F. K. Pittman, 669.

THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

A MEETING of the Society was held at 2.15 p.m., on Wednesday, December 6th, at the Chemical Society's Rooms, Burlington House. The President—Professor W. H. Roberts—was in the chair. The following papers were read and discussed:—

“The Examination of Lard,” by R. W. Sutton, B.Sc., F.I.C., A. Barraclough, B.Sc., F.I.C., R. Mallinder, B.Sc., F.I.C., and O. Hitchen, B.Sc., F.I.C.; and “The Estimation and Examination of 2-Methyl-1:4-naphthoquinone,” by J. L. Pinder, B.Sc., F.I.C., and J. H. Singer, A.I.C., with introductory remarks on the relationship of 2-methyl-1:4-naphthoquinone to Vitamin K, by A. L. Bacharach, M.A., F.I.C.

The following candidates have been elected members of the Society:

Ernest Brown, B.Sc. (Lond.), Chemist with experience of analysis of soaps, essential oils and cosmetics.

Alexander Stuart Robertson Inglis, B.Sc. (Edin.), A.I.C., Assistant Chemist in the County Laboratory, Stafford. (*Through the North of England Section.*)

Samuel Gordon Liversedge, F.I.C., Head of the Analytical Department of Messrs. Howards & Sons, Ilford.

Clifford Hanks Robinson, B.A. (Toronto), F.C.I.C., Chief of the Division of Chemistry, Science Service, Canadian Department of Agriculture, Dominion Agricultural Chemist.

Obituary

WILLIAM THOMAS BURGESS

WILLIAM THOMAS BURGESS, a former member of Council and Vice-President of the Society, died at Bedford Park, London, on June 22nd, 1939, at the age of 78.

A native of Brighton, he received a scientific training there, including chemistry, under the late W. Jago, F.I.C. In 1882 he continued his studies at the Royal College of Science, and, in his first year's course, showed his natural aptitude for physics by taking top place in the examination and the Tyndall Prize. In those days Agriculture was given much prominence in the College, and the Associateship could be obtained in that subject. Burgess was selected from among the most

promising chemistry students to act as a demonstrator in a Summer Course in Agriculture, and his deftness at manipulating apparatus and general resourcefulness in experiments was noted by Professor Percy F. Frankland, who proposed that he should be employed as assistant to his father, Sir Edward Frankland, in a private laboratory where the consulting work of both father and son was done. Burgess's acceptance of this post entailed a stoppage of his work at the College. His fellow assistant for a short time was Frederick (later Sir Frederick) Gowland Hopkins, and, for a few years after, James Kear Colwell, now Public Analyst for Finsbury and elsewhere. From 1884 until the death of Sir Edward, in 1899, Burgess was his sole assistant, and, in this capacity, learned all that was known about water supplies and geological strata throughout the country.

On the death of Sir Edward Frankland it was proposed that his official work as "Water Analyst to the Local Government Board" should be transferred to the Government Laboratory. Dr. Thorpe agreed on condition that Burgess would continue the work as a part-time assistant, and a room was placed at his disposal in which to design and lay out the necessary apparatus. Although the greater part of Burgess's time was taken up with this official work, which consisted mainly in regular testing of the water supplied to London by the eight separate companies, his intimate connection with most of the large water undertakings in the country involved reference to him in the conduct of these, and he continued the consultative work for them, first in the laboratory of Dr. Samuel Rideal, and later in his own home. When the Metropolitan Water Board took over, in 1908, the examination of the London water supply, there was not enough official work in the Government Laboratory to keep Burgess employed, and, wishful of devoting his energies entirely to his private work, he left the Laboratory.

His unrivalled knowledge of the water supplies of this country enabled him to continue a sound practice as water consultant until his death. There were few new installations throughout the land where Burgess was not called in for advice either by the authorities or the water engineers.

He devised a simple colour meter for determining the degree of discoloration in water in terms of the Lovibond scale, and he retained his old love for the Frankland combustion method of determining carbon and nitrogen in water, probably later than any other analyst.

He had been a Member of Council, Vice-President and, temporarily, Honorary Treasurer of the Institute of Chemistry, and did much work for the Glass Research Committee during the last war.

Never robust, Burgess kept alive a boyish interest in every new development in chemical and physical apparatus, and his keen appreciation of the value of any novelty, coupled with helpful advice often wrapped up in an anecdote, endeared him to all with whom he came in contact.

A. MORE

The Detection and Estimation of Benzedrine

By EDWARD T. ILLING, B.Sc., F.I.C.

BENZEDRINE or β -aminopropylbenzene ($C_9H_{11}N$) is chemically allied to ephedrine and adrenaline. It diminishes gastro-intestinal activity, raises the blood pressure and interferes with sleep, and may cause pronounced psychological changes. When applied to mucous membranes it causes contraction of the blood vessels and shrinkage of the membrane, and is used as an inhalant to free congested nasal passages in coryza. It tends to abolish fatigue, especially in depressed patients. A case of acute aplastic anaemia following self-administration of the drug has been recorded¹; 190 mg. were taken in the course of 19 days. Severe cardiovascular collapse occurred the day after the last dose. Gradual recovery ensued.

In connection with a case of sudden death due to haemorrhage in the cranium, an endeavour was made to try to prove the absence of benzedrine in the viscera. Very little information about benzedrine could be found, so that it was necessary to make a study of the substance and its general characteristics with a view to working out a satisfactory method of detection and estimation. This paper sets out some of the results of the study.

Benzedrine is a liquid that is slightly soluble in water, more soluble in alcohol, and readily soluble in acids, ether, amyl alcohol, ethyl acetate and chloroform. It is completely volatile at $100^\circ C.$, and appreciable loss occurs even at low temperatures, as is shown by the following results:

Twenty mg. of benzedrine were dissolved in about 10 ml. of ether, the solvent was evaporated at 70° to $80^\circ C.$, and the residue was dried in a desiccator; weight, 7 mg. \equiv 65 per cent. loss.

A solution of 7 mg. in ether was evaporated at 38° to $40^\circ C.$; residue, 4.2 mg. \equiv 40 per cent. loss.

A solution of 4.2 mg. in ether was evaporated at 33° to $35^\circ C.$; residue, 3.3 mg. \equiv 20 per cent. loss.

The acetate also is volatile:

Twenty mg. of acetate were dissolved in 10 ml. of alcohol, the solvent was evaporated below $40^\circ C.$, and the residue was dried in a desiccator; weight, 12 mg. \equiv 40 per cent. loss.

12 mg. of acetate heated for 15 minutes below $40^\circ C.$; residue, 9.5 mg. \equiv 20 per cent. loss.

9.5 mg. of acetate heated for 15 minutes on the water-bath; residue, 4.5 mg. \equiv 50 per cent. loss.

18.8 mg. of acetate heated for 30 minutes in the oven; residue, 5.8 mg.

Heated for a further 1 hour the residue weighed 0.4 mg.

" " " " 1 " " " " nil.

Hence, 19 mg. of acetate were completely volatilised at $98^\circ C.$ in less than 2 hours.

The hydrochloride, sulphate and tartrate are crystalline salts, are not volatile, and may be heated at $100^\circ C.$ without decomposition. These salts are soluble in water and alcohol. The sulphate is not so soluble as the chloride and tartrate;

for instance, 100 ml. of alcohol dissolved 60 mg. of sulphate at 17.5° C., whereas more than 50 times this amount of hydrochloride dissolved readily in 100 ml. of alcohol at the same temperature. The salts dissolve readily in 50 per cent. alcohol.

ESTIMATION OF BENZEDRINE IN AQUEOUS SOLUTIONS.—The extraction from solution by means of chloroform depends to some extent upon the degree of alkalinity. Sodium bicarbonate was used for the first trials, but it was found that, although the base could be extracted, a large number of single extractions with chloroform was required. For instance, when 20 mg. of benzedrine sulphate were taken and the alkaline solution was about 50 ml. in volume, a series of extractions with 25, 15, 10 and 10 ml. of chloroform extracted 10.5 mg. of benzedrine sulphate

| | | | | | | |
|--|---|-----|---|---|---|---|
| A second similar series of extractions | „ | 7.1 | „ | „ | „ | „ |
| A third | „ | 2.1 | „ | „ | „ | „ |
| <hr/> | | | | | | |
| Total extracted 19.7 „ „ „ „ | | | | | | |

When the solution was made alkaline with sodium hydroxide, one series of extractions with chloroform extracted the whole of the benzedrine sulphate. The method adopted is as follows:—The aqueous (generally acid) solution, containing in 50 to 100 ml. the salt of benzedrine, is neutralised approximately with 10 per cent. sodium hydroxide solution, and 1 ml. is added in excess. The solution is extracted with successive quantities (25, 15, 10 and 10 ml.) of chloroform. Each chloroform extract is washed with the same 5 to 10 ml. of water, this wash water is then shaken with a little chloroform, which is added to the combined extracts, and the whole is washed with 5 ml. of water. The benzedrine is then re-extracted from the chloroform by shaking with four successive portions of 10 ml. of *N*/2 hydrochloric acid. These acid extracts are evaporated to dryness on the water-bath, and finally the residue is dissolved in absolute alcohol, filtered into a tared flask, evaporated to dryness, dried and weighed. In two experiments

- (a) 20.6 mg. of benzedrine hydrochloride gave 20.6 mg. of the hydrochloride.
- (b) 20.0 mg. of benzedrine gave 19.5 mg. of benzedrine.

TEST FOR BENZEDRINE.—Benzedrine gives the general reactions for alkaloids, such as those of Meyer and Dragendorff, but there seemed to be no other test available. However, a new test has been found which is quite delicate and has proved useful in confirming the purity of the residues of hydrochloride. Benzedrine gives a purple colour in the modified Mohler's Test already described by me.² The conditions are exactly the same as those used in the detection of benzoic acid by this test, and the same precautions have to be taken. The test will detect 0.1 mg. of benzedrine, and the colour given by 1.0 mgm. is a convenient one to match in a volume of 15 ml. The purple colour fades quickly on dilution with water, as does also that given by *m*-diamino-benzoic acid; but it has been found that the colour is quite stable if the solution prepared as follows is used as a diluent:—Twenty ml. of conc. sulphuric acid are added to 40 ml. of water containing 2 g. of potassium nitrate, and the liquid is cooled while 200 ml. of water containing 100 ml. of ammonia (sp.gr. 0.880) are added carefully. Finally, 40 ml. of water containing 0.8 gm. of hydroxylamine hydrochloride are added. This solution

may be used for diluting the final 15 ml. of the test solution to 50 ml. or other convenient volume. From 2 to 3 mg. of benzedrine in a volume of 50 ml. give a colour suitable for matching.

Ephedrine and adrenaline under the same conditions give a deep yellow and a slight yellow colour respectively.

DISTILLATION OF BENZEDRINE.—The benzedrine salt is introduced into a 1-litre round-bottomed flask, fitted as is usual for steam-distillation, and 50 ml. of water, 25 g. of salt and sufficient sodium hydroxide to render the liquid alkaline are added. No condenser is necessary, so that the total condensed distillate is small in volume. The distillate is passed through 10 ml. of *N* hydrochloric acid contained in a conical flask (200 ml.), which is connected by means of a glass tube with the outlet of the distillation flask by rubber tubing; the other end of the glass tube passes through a bung in the neck of the conical flask. From this flask the distillate is passed through a further 10 ml. of *N* hydrochloric acid contained in a distillation flask (100 ml.), and the glass tube leading into this flask is drawn out so as to make a slightly smaller orifice; but this must not be too small, otherwise the pressure required to force the steam through the absorbers would be too great. Sufficient water to form a seal is added to the 10 ml. of hydrochloric acid in each flask, and the steam finally escapes through the side tube of the last flask.

The distillation is continued for about 30 minutes, and the main flask is heated in such a way that a small amount of salt remains undissolved. At the end of the distillation the flame is kept beneath the flask while the steam is stopped and the connection removed, then the outlet of the main flask is disconnected from the conical flask. The combined distillates are evaporated to about 50 ml., transferred to a separator, made alkaline with sodium hydroxide, and extracted as described above.

In two test experiments, in which 20 mg. of benzedrine were taken, 18.7 and 18.9 mg. were recovered.

EXTRACTION FROM VISCERA.—Benzedrine may be separated from viscera by the Stas-Otto process, it being borne in mind that the acetate is volatile, so that if acidification is required at any stage before or during evaporation, tartaric acid should be used. The benzedrine is extracted from the alkaline solution with chloroform. It is suggested that a portion of the chloroform extract should be extracted with hydrochloric acid, as described above, to prove the presence or absence of benzedrine. It is also preferable to render the solution alkaline with sodium bicarbonate when dealing with an unknown specimen and to make further extractions with chloroform.

In one experiment, 3.2 mg. of benzedrine hydrochloride were added to 40 g. of stomach and contents, and 2.8 mg. of hydrochloride were recovered.

Distillation.—The distillation of alkaline mixtures of viscera is practically impossible owing to the excessive frothing. Attempts were made to form the acetate and to distil from an acetic acid solution, but without success. No trace of benzedrine could be detected in the residues.

EXTRACTION FROM URINE.—The following experiment is typical:—The benzedrine (18.9 mg.), as tartrate, was added to 50 ml. of urine, and the mixture was steam-distilled for 1 hour, as previously described. From the distillate 24.5 mg.

of impure hydrochloride (= 19.3 mg. of impure benzedrine) were obtained, and Mohler's test showed that the residue contained 18.0 mg. of pure benzedrine.

BENZEDRINE TABLETS.—Each tablet is stated to contain 5 mg. of β -amino-propyl benzene sulphate. It was found that absolute alcohol did not extract all the benzedrine sulphate (actually only 3.75 mg. per tablet were obtained by extracting 2 tablets), but if the sulphate is converted into the hydrochloride a good extraction is possible.

Thus, 0.892 g. of crushed tablets contained in a 100-ml. conical flask was mixed with a little solid barium chloride and 1 ml. of *N* hydrochloric acid. Twenty-five ml. of absolute alcohol were added, and the mixture was heated on the water-bath for a few minutes. The alcohol was filtered, and the residue was washed with absolute alcohol. All the alcohol was evaporated from the filtrate without allowing the filtrate to evaporate to dryness. The liquid was filtered into a separator, the filter was washed with acidified water, the filtrate was approximately neutralised with 10 per cent. sodium hydroxide solution, and 1 ml. was added in excess. The extractions were made with chloroform, as already described. The amount of hydrochloride obtained was 12.2 mg., which is equivalent to 4.8 mg. of benzedrine sulphate per tablet.

Alcohol of 50 per cent. strength will extract the sulphate completely, and this method is quite satisfactory. The tablets are extracted with 25 ml. of 50 per cent. alcohol and the process described above is carried out, but omitting the treatments with barium chloride and hydrochloric acid. The alcoholic filtrate should be made slightly acid with 2 to 3 ml. of *N*/10 hydrochloric acid. [No more acid should be added during the heating to drive off the alcohol, for if even 1 ml. of 10 per cent. hydrochloric acid is added, no benzedrine will be extracted; moreover, attempts have been made to prepare the solution for extraction by heating the tablets for 1 hour on the water-bath with 25 ml. of water containing 5 ml. of conc. hydrochloric acid, but no trace of benzedrine was obtained in the final residue.]

This process gave 4.9 mg. of benzedrine sulphate per tablet, and Mohler's test showed the residue to be pure.

Distillation.—For this process 1.4162 g. of tablets were taken and the benzedrine was distilled from an alkaline salt solution as already described; 16.8 mg. of benzedrine hydrochloride were obtained, which is equivalent to 4.2 mg. of benzedrine sulphate per tablet. It was found necessary to prolong the distillation to 1 hour, as the benzedrine evidently distils at a slower rate from this solution containing the tablets. In a second experiment, 0.8812 g. of tablets was taken, and the distillation was carried out for 1 hour; 11.4 mg. of hydrochloride were obtained, which is equivalent to 4.55 mg. of benzedrine sulphate per tablet. When 20 mg. of benzedrine were distilled only 18.8 mg. were recovered and, if this is made the basis of a correction, the 4.55 mg. are equivalent to 4.9 mg. of benzedrine sulphate per tablet.

SULPHATE.—One or two tablets, or a portion of crushed and mixed tablets, were heated for 1 hour on the water-bath with 25 ml. of water and 5 ml. of conc. hydrochloric acid. The solution was filtered, the residue was washed thoroughly, and the sulphate was estimated in the filtrate.

The barium sulphate is precipitated slowly and must be allowed to stand some hours, preferably overnight. The precipitation is complete in 3 hours if the tablets are dissolved in 25 ml. of water containing 2 ml. of 10 per cent. sodium hydroxide by heating on the water-bath and then adding 5 ml. of concentrated hydrochloric acid and proceeding as above.

The results of the estimation of benzedrine in the tablets are collected together in the following table:

| | | Method | | | |
|--------------------------------|---|--------------------------|---------------------------------|-------------------------------|---|
| Sample taken | | 50 per cent. alcohol mg. | Steam distillation (1 hour) mg. | Determination of sulphate mg. | BaCl ₂ acid and absolute alcohol mg. |
| Benzedrine sulphate per tablet | { one or more separate tablets } | 4.9 | — | 4.5 | — |
| | | 4.5 | — | 5.3 | — |
| | { 20 tablets crushed and portions taken } | 4.9 | 4.5 | 4.5 | 4.8 |
| | | 5.2 | (corrected figure 4.9) | 5.3 | 5.1 |
| | | — | — | 5.4 | — |
| | | — | — | — | — |

In conclusion, I wish to thank Mr. D. R. Wood, F.I.C., for his interest and criticism, and Mr. E. G. Whittle, B.Sc., A.I.C., for his assistance in carrying out some of the determinations.

REFERENCES

1. I. J. Davies, *Brit. Med. J.*, 1937, II, 615.
2. E. T. Illing, *ANALYST*, 1932, 57, 225.

THE COUNTY LABORATORIES
TAUNTON, SOMERSET
July 7th, 1939

Examination and Determination of 2-Methyl-1:4-Naphthoquinone

By J. L. PINDER, B.Sc., F.I.C., AND J. H. SINGER, A.I.C.

(Read at the Meeting, December 6, 1939)

INTRODUCTION.—The independent discovery, by Dam and by Almquist and their collaborators,^{1,2,3,4} of a vitamin necessary for maintenance of normal blood-clotting behaviour in the young chick at first roused interest only among those directly concerned with poultry husbandry. Later investigations, stimulated by Dam's own first clinical trials,⁵ pointed to certain important therapeutic uses of the naturally occurring active substance, whose richest sources appear to be certain green leaves, especially those of the chestnut. It is also synthesised by certain bacteria. Intensive research during the last three years has now established

the following facts about the vitamin, which has been called vitamin K ("Koagulationsvitamin"):

- (1) It exists naturally in at least two forms; one is the phytyl derivative of 2-methyl-1:4-naphthoquinone and the other is derived from the same quinone and another alcohol, possibly farnesol. The two forms have somewhat different biological activities on young growing chicks.
- (2) The blood-clotting time of young chicks deprived of the vitamin rises to many times its normal value, and the animal eventually succumbs to a multiple haemorrhagic condition.
- (3) There is no direct evidence for the existence or experimental production of vitamin K deficiency in any animals besides chickens and one or two other species of poultry.
- (4) Attempts to use vitamin K preparations on human subjects suffering from obstructive jaundice and certain other conditions associated with prolonged blood-clotting times (specifically with a reduction in plasma prothrombin and an increased "prothrombin time") have given encouraging results. Similar results, though so far only a few, have been claimed from treatment with the synthetic vitamin (phytyl derivative).
- (5) 2-Methyl-1:4-naphthoquinone appears to exert on the chick qualitatively and quantitatively the same action as the "natural" forms of vitamin K.

From this it is but a short step to the introduction of the methyl naphthoquinone itself as a therapeutic agent. Evidence is now available of its efficacy.⁶ It is probable, therefore, that a method for the detection and estimation of this particular naphthoquinone will be required in the assay and in control of the purity of the substance in bulk and to estimate it in the various preparations in which it may be used; the work described below was undertaken to provide such a method.

EXAMINATION AND DETERMINATION

(a) BULK SUPPLIES.—A common method of preparing 2-methyl-1:4-naphthoquinone is by oxidising methyl-naphthalene with chromic acid, followed by recrystallisation. The quinone itself is decomposed in sunlight, changing from a lemon-yellow colour to a light brown. The end-product is thought to be 2,3,2',3'-tetrahydro-2,3'-dimethyl-2,3,2',3'-binaphthalene-1,4,1',4'-tetrone.⁷ A method for assaying the pure substance must permit estimation of the quinone in presence of possible traces of unchanged methyl-naphthalene, and also of traces of the decomposition product. The melting-point (106° C.) is a useful criterion of purity; unchanged methyl-naphthalene has m.p. 32° C., and the m.p. of the tetrone is given as 235° C.⁷ The presence of either of these substances would therefore have a considerable effect. Tests must also be applied for excess ash, and for traces of chromium and of solvent. The compound should be stored in brown bottles in the dark, and all tests should be made in the absence of direct sunlight.

The quinone can be estimated by means of its absorption spectrum.

The following chemical assay method was devised after preliminary work had established that under suitable conditions the quinone could be quantitatively

reduced to hydroquinone by means of titanous chloride, the end-point of the reduction being shown by the use of an internal oxidation-reduction indicator.

Determination.—Weigh out accurately about 0.2 g. of the substance, and transfer it to a 200-ml. conical flask. Dissolve it in a mixture of 10 ml. of alcohol (industrial methylated spirit) and 15 ml. of glacial acetic acid. Add 4 g. of anhydrous sodium carbonate and 25 ml. of a 10 per cent. aqueous solution of sodium potassium tartrate (Rochelle salt). Observe these quantities of carbonate and tartrate, as they control the pH of the solution and prevent its becoming too acid during the course of the titration. Titrate with an approximately *N*/10 solution of titanous chloride in *N* hydrochloric acid, using 3 drops of a 0.1 per cent. aqueous solution of potassium indigo-disulphonate as internal oxidation-reduction indicator; a sharp end-point is obtained, the blue dye being changed to the colourless leuco base by the addition of one drop of the titanous chloride solution in excess. Phenosafranine is also a suitable indicator. During the titration pass a steady stream of carbon dioxide through the solution.

The titanous chloride solution is stored under hydrogen or carbon dioxide in the usual way, and is periodically standardised against a ferric iron solution of known strength (1 ml. of *N*/10 titanous chloride \equiv 0.0086 g. $C_{11}H_8O_2$). Suggested limit, not less than 98.5 per cent.

Appearance and Odour.—A lemon-yellow crystalline powder with a faint but characteristic odour. No odour of methyl-naphthalene should be discernible.

Ash.—One g. is ashed in a platinum dish over a Bunsen flame, and finally in the muffle furnace. Suggested limit, not more than 0.15 per cent.

Chromium.—The ash is fused with a small quantity of a mixture of sodium and potassium carbonates and sodium peroxide, the cooled melt is dissolved in water, and the solution is acidified with dilute sulphuric acid and made up to 50 ml. An aliquot part is transferred to a test-tube and diluted with water, and a few drops of diphenyl-carbazide reagent (0.2 per cent. in a mixture of one part of glacial acetic acid and nine parts of alcohol) are added. The violet solution is made up to about 10 ml. and its colour is compared with those produced from known quantities of a standard solution of chromium (0.283 g. of $K_2Cr_2O_7$ in 1 litre: 1 ml. \equiv 0.1 mg. Cr. Suitable dilutions of this stock solution are made as required). Suggested limit, not more than 0.01 per cent.

Loss in Vacuum Desiccator.—One g. is weighed into a small tared glass crystallising dish, which is transferred to a vacuum desiccator. The dish is covered with an inverted tin (to exclude light) and dried to constant weight. Suggested limit, not more than 0.2 per cent.

Melting-point.—This should be 104–106° C.

(b) **COLORIMETRIC ESTIMATION OF SMALL QUANTITIES.**—A reliable method for the estimation of small quantities of 2-methyl-1:4-naphthoquinone in tablets and ampoules was also required. A search of the literature revealed several colour tests for quinones; of these, three were investigated.

- (1) Raudnitz and Puluj⁸ describe a reaction between aldehydes and quinones in acetic acid solution in presence of hydrochloric acid. Many quinones are stated to yield coloured products. The reaction was tried with a variety of aromatic and aliphatic aldehydes and a representative selection

of quinones; in some instances colours were obtained, but further investigation showed that the same colour could be produced by the aldehyde with hydrochloric acid alone. No initial success was forthcoming, and the method was abandoned.

- (2) The colour reaction with sodium methylate, proposed by Almquist and Klose^{9,10} for natural concentrates of vitamin K, was tried; the results with 2-methyl-1:4-naphthoquinone were inconclusive.
- (3) Craven¹¹ describes colours developing on the addition of ethyl cyanacetate to a solution of a quinone in equal volumes of ammonia (sp.gr. 0.880) and alcohol. This test was tried on a number of quinones; benzoquinone, sodium β -naphthoquinone sulphonate and 2-methyl-1:4-naphthoquinone all yielded strong colours. The mixed reagents alone developed no colour, and the test was therefore investigated at some length.

Preliminary experiments showed that 0.1 mg. of methyl-naphthoquinone in alcoholic solution gave a detectable violet colour in 3 ml. of the ammonia and alcohol mixture with 3 drops of ethyl cyanacetate, whilst 0.5 mg. gave a good colour. The coloured solution on dilution with water underwent little change in tint; it was also fairly stable to dilute alkali. Addition of strong alkali (6 *N* potassium hydroxide solution) produced a stable yellow colour. The original violet colour was destroyed by acid, and was not extracted by ether. The violet colour faded fairly rapidly and was therefore unsuitable for colorimetric estimations, but the yellow colour with alkali gave more promise. Comparison between the colours given by the same quantity of quinone, using in the Hilger (Spekker) absorptiometer the filters giving the maximum readings, showed that the yellow alkali colour was more intense than the original violet. Efforts were then made to find the best conditions for maximum development and stability of the yellow colour, combined with consistency of results.

In the original method 3 ml. of the alcohol and ammonia solution were added to the alcoholic solution of the quinone, followed by 3 drops of ethyl cyanacetate and then 5 to 6 ml. of 6 *N* aqueous potassium hydroxide solution. The boiling-tube was then warmed over a micro burner to incipient boiling of the liquid. The colours produced were not strictly reproducible; this was found to be due to two causes:

- (a) The initial violet colour, as already indicated, fades fairly rapidly; if the alkali is added shortly after the ethyl cyanacetate, a more intense yellow is obtained than if the alkali is added after the violet has begun to fade.
- (b) The yellow colour itself is not completely stable, and is more rapidly destroyed on warming than on standing at room temperature. It was found that maximum colour development and reliable duplication of results were obtained by adding the alkali within half a minute of the cyanacetate and by allowing the yellow colour to develop in the cold for about 15 minutes. The solutions are then made up to 50 ml. with water and the absorption intensity is measured.

The effect of the volume of the alcoholic solution of quinone was also investigated, and it was found that, under the conditions outlined above, 0.5 mg. of quinone

gave within experimental error the same reading when dissolved in amounts ranging from 1 to 6 ml. of alcohol.

The following details of the test are therefore proposed:—To 1 to 5 ml. of an alcoholic solution containing 0.4 to 0.8 mg. of the quinone, add 3 ml. of a mixture of equal volumes of alcohol (industrial methylated spirit) and ammonia (sp.gr. 0.880). Add 3 to 5 drops of ethyl cyanacetate and allow the mixture to stand for exactly half a minute. Add 5 ml. of 6 *N* potassium hydroxide solution, mix, and leave for 15 minutes, preferably out of direct sunlight. Dilute to 50 ml. and measure the absorption in the Hilger (Spekker) absorptiometer, using the light blue No. 6 filter. Read off the quantity of quinone from a previously prepared graph.

Application of the Method.—The method may be applied to tablets and pills; these are ground to powder in a mortar, and extracted in the cold with alcohol, the volume being finally adjusted in a graduated flask, and a suitable aliquot part is pipetted out for assay as described above. It may also be applied to solutions in arachis or other oils by shaking a suitable weighed quantity with alcohol, and proceeding with the determination on the resulting emulsion; the alcohol extracts the quinone from the oily solution. Any undissolved oil may be removed after the final dilution with water by shaking with ether, in which the yellow colour is insoluble.

RESULTS

The following table summarises a typical series of results:

| Sample | m.p. ° C. | Loss in vacuum Per Cent. | Ash Per Cent. | Chromium Per Cent. | Quinone | | Spectrographic examination | | Quinone calculated from absorption band at 250m μ Per Cent. |
|--------|--------------|-----------------------------------|---------------------|--------------------------|----------------------------|------------------------------|-------------------------------|------------------|---|
| | | | | | Volumetric Per Cent. | Colorimetric Per Cent. | F _{100m} % | | |
| | | | | | | | 331m μ | 250m μ | |
| A | 103-105 | 0.18 | 0.18 | 0.06 | 97.7 | — | 143 | 1110 | 95 |
| B | 104-106 | 0.19 | 0.10 | 0.005 | 99.1 | — | 155 | 1160 | 99 |
| C | 104-106 | 0.19 | 0.10 | 0.005 | 98.8 | — | 158 | 1160 | 99 |
| D | 102-106 | — | — | — | 93.4 | — | 145 | 1090 | 93 |
| E | 125-220 | — | — | — | 18.3 | 19 | 29.5 | band obscured | 19* |

* Calculated from the absorption at 331 m μ .

Special interest attaches to sample E. Part of sample A was transferred to a small glass specimen tube and exposed to the light under laboratory conditions for a period of about six weeks. During this period its colour changed to a light brown. As indicated in the table, it melted over a very wide range. The various assay processes gave results in agreement, indicating that all three methods measure the quinone and are uninfluenced by the presence of the decomposition products.

SUMMARY.—1. A volumetric method for the assay for 2-methyl-1:4-naphthoquinone has been devised.

2. Control tests for likely impurities are described.

3. The spectrographic absorption has been measured.

4. A colorimetric method, capable of application to tablets and ampoules, has been developed for estimating quantities of the order of 0.5 mg.

5. A summary of results obtained by these tests is given.

We have to acknowledge the encouragement and help of Mr. A. F. Lerrigo, B.Sc., F.I.C., during both the execution and the presentation of our experimental work; the co-operation of Miss B. E. Stern, B.Sc., in carrying out all the spectroscopic observations involved; the assistance of Mr. A. L. Bacharach, M.A., F.I.C., in supplying the historical and biochemical data of the introductory paragraphs; and permission to publish this paper given us by the Directors of Glaxo Laboratories Ltd., Greenford.

REFERENCES

1. H. Dam, *Nature*, 1935, **135**, 652.
2. ——— *Biochem. J.*, 1935, **29**, 1273.
3. H. J. Almquist and E. L. R. Stokstad, *Nature*, 1935, **136**, 31.
4. ——— *J. Biol. Chem.*, 1935, **111**, 105.
5. H. Dam and J. Glavind, *Lancet*, 1938, **234**, 720 (Vol. 1).
6. J. M. Macfie, A. L. Bacharach and M. R. A. Chance, *Brit. Med. J.*, 1939, **II**, 1220.
7. J. Madinavietia, *Anales. Soc. españ. fis. quim.*, 1933, **31**, 750-9.
8. H. Raudnitz and G. Pulu, *Ber.*, 1931, **64B**, 2212-6.
9. H. J. Almquist and A. A. Klose, *J. Amer. Chem. Soc.*, 1939, **61**, 1611.
10. ——— *ibid.*, 1939, **61**, 1923.
11. R. Craven, *J. Chem. Soc.*, 1931, 1605-6.

ANALYTICAL DEPARTMENT
GLAXO LABORATORIES, LTD.
GREENFORD
MIDDLESEX

October, 1939

DISCUSSION

Mr. L. EYNON asked if 2-methyl-1:4-naphthoquinone replaced not only vitamin K₁ but vitamin K₂.

Dr. H. E. COX said that he found the paper most interesting because it gave a new importance to the naphthoquinones and re-opened the puzzle of the function of substituted naphthoquinones in plant life. He thought that experiments on walnuts, which contain much juglone (5:hydroxynaphthoquinone), would be worth making. With regard to the titration of methyl naphthoquinone it was interesting to note the effect of the methyl group in reducing the activity of the quinone. He had found that the 2-hydroxy compound was so active a reducing agent that it could not be titrated with titanous chloride and had had to devise a special method for its titration (ANALYST, 1938, **63**, 399); when the hydroxyl group was replaced by -CH₃ the reducing power was much diminished, though the resulting product was so much more sensitive to light. He hoped the authors would continue their work on other naphthoquinone derivatives.

Mrs. TRITTON asked whether 2-methyl-1:4-naphthoquinone was as stable as vitamin K. She also asked how the tests were made. Was the substance given by the mouth or by intravenous injection?

Mr. PINDER, replying to Dr. Cox, said that quite a number of batches of 2-methyl-1:4-naphthoquinone had been estimated, by means of titanous chloride, with very satisfactory results. He had recently had occasion to try the method on a hydroxyquinone, but the results were unsatisfactory. He had come to the conclusion that the presence of hydroxy groups rendered the method useless.

Mr. BACHARACH thought that methyl-naphthoquinone would replace both vitamin K_1 and vitamin K_2 for all practical purposes. Its stability was about the same as that of vitamin K_1 , though it was rather less photo-labile. It was possible that vitamin K_1 might be associated in plant tissues with other naphthoquinone derivatives. This clue to likely sources was, however, not available to Dam and the other earlier investigators—on whose work his (Mr. Bacharach's) table of vitamin K distribution had been based—because the naphthoquinonoid structure of the vitamin had not at that time been made clear. Almost all of the tests on chicks had been carried out by the oral route. In the clinical use of vitamin K_1 or 2-methyl-1:4-naphthoquinone, there were three possible routes. First the oral, less practical, because it almost invariably involved simultaneous administration of bile salts; second, intravenous, in diluted aqueous alkalis solution, where this can be prepared; third, intramuscular or subcutaneous, in an oil solution.

The Assay of Mercury

By J. SANDILANDS, F.I.C.

ALTHOUGH there are numerous methods for the assay of mercury, it is noteworthy that, except in those of Hempel,¹ Personne² and a few others, the formation of iodide as a feature of the method has been neglected. This seems strange when one considers the ease with which mercury and iodine combine to form a highly insoluble compound, somewhat refractory to ordinary reagents, and volatile, though not so much so as mercury itself.³ The readiness with which mercuric iodide forms double salts and complexes may be a reason for this neglect, and, with the exception of the methods in which the element itself is produced in the assay, in nearly every instance the procedure involves the formation of a mercuric salt in some liquid medium from which the sulphide is precipitated and weighed. When the conversion of the material into mercuric salt can be carried out—a tedious process when much organic matter is present—the thiocyanate method,⁴ analogous to that of Volhard for halides, is available. This method, however, has distinct limitations.⁵ B. Rose⁶ found it necessary in his dry assay of mercury to heat the material with quicklime and a finely divided metal, but others have shown that the complete decomposition of all mercury compounds cannot be effected in this way. In consequence of this, such modifications of the method as those of Chism,⁷ Holloway (Eschka),⁸ Meharg,⁹ and H. ter Meulen¹⁰ have been devised.

It should be noted that all wet methods in which prolonged treatment with acid is essential for decomposition are liable to loss, owing to the volatility of the salt from a *dry* part of the reaction vessel. Apparently there is less loss in an acid than in a neutral medium, as is shown in Table I. This gives the results obtained on keeping an aqueous solution of mercuric chloride at a definite temperature for a given time as compared with those obtained with a similar solution in 5 per cent. hydrochloric acid. The solutions were placed in beakers with the same area exposed to the atmosphere. They were heated electrically on a hot plate, and draughts were excluded by having asbestos screens round the

vessels. The temperature selected was 85° C., the duration of each test was 8 hours, and periodically solvent was added to replace any evaporation loss. The results agree closely with those obtained by Sulc,¹¹ Estere,¹² and Lehmann.¹³

TABLE I

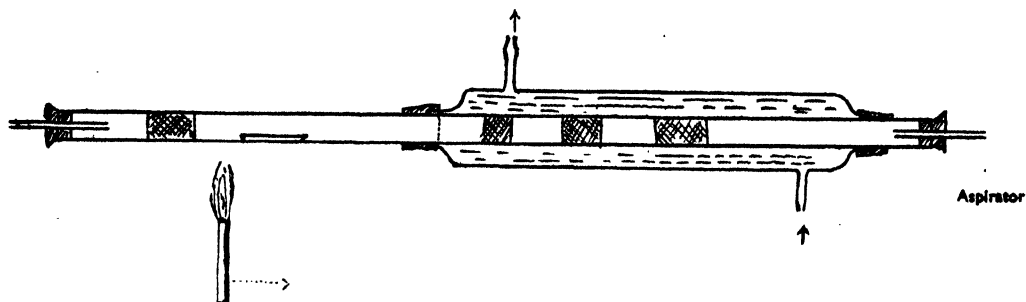
Aqueous Solution of Mercuric Chloride kept at 85° C. for 8 hours.

| Pure mercuric chloride taken g. | Mercuric sulphide pptd. | Mercuric chloride equivalent to mercuric sulphide | Difference g. |
|------------------------------------|-------------------------|---|------------------|
| 0.5013 | 0.3908 | 0.4681 | -0.0332 |
| 0.3274 | 0.2544 | 0.3046 | -0.0228 |
| 0.3833 | 0.2985 | 0.3578 | -0.0256 |
| 0.2558 | 0.1977 | 0.2367 | -0.0191 |
| 0.2810 | 0.2164 | 0.2593 | -0.0262 |

Five per cent. Hydrochloric Acid Solution of Mercuric Chloride under same conditions

| | | | |
|--------|--------|--------|---------|
| 0.2873 | 0.2240 | 0.2856 | -0.0015 |
| 0.5094 | 0.4365 | 0.5109 | +0.0015 |
| 0.2654 | 0.2258 | 0.2642 | -0.0012 |
| 0.4048 | 0.3445 | 0.4032 | -0.0016 |
| 0.2775 | 0.2380 | 0.2780 | +0.0005 |
| 0.7058 | 0.6040 | 0.7065 | +0.0007 |
| 0.3928 | 0.3367 | 0.3941 | +0.0013 |

The original purpose of this work was to endeavour to get mercuric iodide directly from a compound, but the difficulty of collecting the product in a confined space proved too great an obstacle. Instead, the iodide was dissolved in sodium thiosulphate solution,¹⁴ excess of thiosulphate was removed by addition of a little iodine, and mercuric sulphide was precipitated from the solution in the normal way. The first series of experiments was conducted on a specimen of cinnabar known to contain 57.5 per cent. of mercury. A quantity of ore was weighed into



a combustion boat, mixed with four or five times its weight of iodine, and introduced into a combustion tube, almost half of which formed the inner tube of a water-cooled condenser (Fig. 1). Within the cooled portion, and acting as baffles, were some plugs of asbestos fibre, while a tighter plug was situated at the inlet end of the tube as a safeguard against back-firing. Later, glass beads were substituted for the asbestos plugs. A gentle stream of air was drawn through the apparatus by means of an aspirator, but as the experiments progressed it was found more

advantageous to attach the exit tube to a controlled suction pump. During the tests on cinnabar, the results of which are given in Table II, it was noticed that the condensed product varied in colour.

TABLE II

| Ore taken g. | Mercuric sulphide (supposed) obtained g. | Mercury Per Cent. |
|-----------------|--|----------------------|
| 0.3851 | 0.3077 | 68.86 |
| 0.2184 | 0.1630 | 63.87 |
| 0.3966 | 0.3054 | 66.39 |
| 0.2877 | 0.2018 | 60.48 |
| 0.4145 | 0.3115 | 64.75 |

To elucidate this irregularity several tests were made without the use of baffles, and some of the solid was obtained. It was washed with carbon disulphide and then with acetone, and the residue was found to contain mercury, iodine and sulphur apparently in combination. The peculiar results thus seem to be due to the formation of thio-iodide; there are a number of such compounds.¹⁵

Experiments to eliminate the sulphur by using iodine in admixture with potassium chlorate in the combustion boat were usually unsuccessful, owing to the speed of the reaction and the consequent loss of the products. With potassium nitrate, in place of the chlorate, the reaction was also vigorous, while attempts to control the rate of reaction by the addition of a non-reactive diluent, such as sand, were no more hopeful. What did emerge from the tests was that a brighter and more uniformly coloured product was formed.

PROCEDURE WITH ORES.—In order to supply iodine and, at the same time, oxygen for the sulphur, iodic acid was next tried. After some preliminary tests the following method was adopted:—The mixture for the combustion boat consisted of one part of ore to between two and three parts by weight of partly dehydrated iodic acid. By leaving the acid in an oven at 95° C. for some time before use the amount of water formed in the reaction later on was reduced; also, a better mixture of the ingredients was obtained. After the heating, when all signs of reaction had ceased, the excess of iodine was removed, together with the red mercuric iodide, by solution in the minimum amount of sodium thiosulphate solution, and the mercury was precipitated with hydrogen sulphide. The results in Table III were obtained with the ore containing 57.5 per cent. of mercury.

TABLE III

| Ore taken g. | Mercuric sulphide from HgI ₂ g. | Mercury Per Cent. |
|-----------------|--|----------------------|
| 0.2101 | 0.1390 | 57.03 |
| 0.3218 | 0.2144 | 57.42 |
| 0.3762 | 0.2579 | 57.7 |
| 0.5466 | 0.3672 | 57.9 |
| 0.3445 | 0.2294 | 57.39 |
| 0.3510 | 0.2342 | 57.5 |
| 0.3813 | 0.2548 | 57.61 |
| 0.2375 | 0.1591 | 57.72 |
| 0.4103 | 0.2767 | 57.85 |
| 0.3758 | 0.2509 | 57.57 |

TABLE IV

No. 1. *Merthiolate (sodium mercurithiosalicylate)*.¹⁶

| Amount taken g. | Mercuric sulphide obtained g. | Mercury Per Cent. |
|--------------------|-------------------------------------|----------------------|
| 0.2579 | 0.1470 | 49.14 |
| 0.2205 | 0.1272 | 49.73 |
| 0.2856 | 0.1629 | 49.17 |
| 0.3014 | 0.1728 | 49.28 |
| 0.2723 | 0.1564 | 49.46 |
| 0.3115 | 0.1780 | 49.27 |
| 0.2422 | 0.1384 | 49.28 |
| Theoretical figure | .. | 49.58 |
| By wet process | .. | 49.36 |

No. 2. *Sublamin (ethylenediamine mercuric sulphate)*.¹⁷

| | | |
|--------------------|--------|-------|
| 0.5015 | 0.2543 | 43.7 |
| 0.2514 | 0.1282 | 43.95 |
| 0.2856 | 0.1465 | 43.92 |
| 0.3237 | 0.1633 | 43.5 |
| 0.2766 | 0.1402 | 43.69 |
| 0.3058 | 0.1556 | 43.86 |
| Theoretical figure | .. | 43.7 |
| By wet process | .. | 43.66 |

No. 3. *Planochrome* (a variety of mercurochrome, 220; hydroxymercuridibromofluorescein)*.¹⁸

| | | |
|--------|--------|-------|
| 1.1549 | 0.2850 | 21.28 |
| 0.8457 | 0.2095 | 21.4 |
| 1.1204 | 0.2767 | 21.3 |
| 0.8846 | 0.2181 | 21.25 |
| 0.7533 | 0.1873 | 21.44 |
| 0.7247 | 0.1780 | 21.42 |

No. 4. *Merfenil† (a phenyl mercuric nitrate)*.¹⁹

| | | |
|--------|--------|-------|
| 0.3415 | 0.2427 | 61.28 |
| 0.2853 | 0.2022 | 61.1 |
| 0.3288 | 0.2341 | 61.48 |
| 0.2510 | 0.1780 | 61.19 |
| 0.3205 | 0.2279 | 61.32 |

No. 5. *Merfenil 002.*

| | | |
|---------------------------|--------|-------|
| 0.2376 | 0.1732 | 62.52 |
| 0.2547 | 0.1843 | 62.4 |
| 0.2283 | 0.1655 | 62.5 |
| 0.3145 | 0.2272 | 62.29 |
| 0.3572 | 0.2593 | 62.6 |
| Figure supplied by makers | | 62.2 |
| By wet process (average) | | 62.34 |

* This compound is liable to contain water. There appear to be several closely related compounds. A wet assay of the sample gave 21.72 per cent. of mercury.

† The sample was supplied by Messrs. May and Baker, Dagenham; its mercury content was stated to be 61.2 per cent.

As the results were satisfactory and were obtained in much less time than by the wet process, the possibility of applying the method to organic mercurials was next investigated.

Since many of the organic mercury derivatives exhibit varying degrees of resistance to treatment during assay, material was selected to embrace various linkages, including the C-Hg type, which is recognised to be the most resistant.

PROCEDURE WITH MERCURIALS.— A quantity of the substance was weighed and mixed on glazed paper with two to four times its weight of dried (95° C.) iodic acid. The mixture was carefully transferred to the boat, and this was introduced into the tube, the outlet of which was connected with the pump by rubber tubing with a screw clip. Heat was applied very gently at the beginning, and was reduced at the first sign of reaction taking place. A bright red deposit, together with some iodine, was seen in the cooled part of the tube. When all reaction had ceased and no further change appeared to take place on prolonged heating, the tube was allowed to cool, and the cooling water was run off. One end of the reaction tube was closed with a small rubber stopper, and two or three crystals of sodium thiosulphate were introduced at the open end. This was followed by a few ml. of water, after which a second rubber stopper was used to close the tube, and the solution was tilted backwards and forwards to dissolve the iodine and the iodide. In some of the tests a few more crystals of thiosulphate had to be added, and in one experiment a little alcohol was introduced, as the residue seemed to be oily. The *cold* solution was filtered, and the filtrate and washings were treated with a few ml. of iodine (alcoholic) until faint reactivity to starch paper was noted; this was to remove excess of thiosulphate. The liquid was then trebled in volume by adding water acidified with dilute hydrochloric acid and slightly warmed, and hydrogen sulphide was passed in. The mercuric sulphide showed the usual variations in colour, and the black precipitate was finally obtained. This was filtered off in a sintered Jena crucible, washed, purified, dried and weighed. Typical results are given in Table IV.

The method here described for the assay of mercury is more rapid than, and as accurate as, the digestion methods. The substance that gave the greatest difficulty was Planochrome, the assay of which by the wet digestion method took almost eight hours as against two hours ten minutes by the iodic acid process.

REFERENCES

1. W. Hempel, *Annalen*, 1859, 110, 176.
2. J. Personne, *J. Pharm. Chim.*, 1863, 43, 477; *Compt. rend.*, 1863, 56, 951.
3. J. W. Mellor, "*Treatise on Inorganic Chemistry*," Vol. IV, 1923, 909.
4. *J. prakt. Chem.*, 117, 217; *Annalen*, 1878, 190, 57.
5. I. M. Kolthoff and N. H. Furman, "*Volumetric Analysis*," II, 245.
6. B. Rose, *Eng. Min. J.*, 66, 480.
7. G. T. Holloway, *ANALYST*, 1916, 31, 66.
8. *Chem. Engineering*, 4, 169.
9. F. C. Whitmore, "*Organic Compds. of Mercury*," New York, 1921, p. 365.
10. H. ter Meulen, *Rec. Trav. Chim. Pays-bas*, 1926, 368; *Abst.*, *ANALYST*, 1926, 51, 422.
11. O. Sulc, *Z. anorg. Chem.*, 1900, 25, 400.
12. E. Esteve, *Chem. Ztg.*, 1911, 35, 1152.
13. F. Lehmann, *Pharm. Ztg.*, 45, 209, 626.
14. J. M. Eder and G. Utm, *Monatsh.*, 1882, 3, 197.
15. J. W. Mellor, "*Treatise on Inorganic Chemistry*," IV, 1923, 961.
16. T. E. Thorpe, "*Dict. Appl. Chem.*," II, Suppl., 1935, 493.
17. *Gazz. Chim. Ital.*, 1909, 39, 143; *Z. anorg. Chem.*, 1902, 32, 231.
18. *J. Chem. Soc.*, 1932, 724; *J. Amer. Chem. Soc.*, 1927, 49, 1227; *id.*, 1920, 42, 3355; *Quart. J. Pharm.*, 1932, 5, 1.
19. *Ber.*, 1897, 30, 510; 1914, 47, 2751.

The Potentiometric Titration of Glucose with Alkaline Tartrate Solutions of Copper, including Fehling's Solution

By H. T. S. BRITTON, D.Sc., F.I.C., AND LESLIE PHILLIPS, M.Sc., A.I.C.

THE use of alkaline tartrate solutions of copper for the determination of reducing sugars was first suggested by Barreswill in 1844, and the method was subsequently elaborated by Fehling.¹ For the purpose, a solution was recommended containing 69.28 g. of $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, 346 g. of Rochelle salt and 130 g. of sodium hydroxide in 2 litres, *i.e.* a solution containing these substances in the molecular ratio of 1:4.6:10.8, respectively. The choice of such a ratio appears to have been entirely fortuitous, the probable reason being that, in the presence of such an amount of tartrate, cupric oxide is maintained in alkaline solution. It is under these conditions that cupric oxide becomes most readily susceptible to reduction, and will in consequence oxidise glucose and other reducing sugars. Unfortunately, the oxidation of glucose is not governed by any definite stoichiometric reaction; hence the statement of Fehling, that exactly five molecules of copper oxide are necessary to oxidise one molecule of glucose, has been shown by several workers to be erroneous. The precise amount of cupric oxide required depends on experimental conditions, particularly those of concentration and *pH*. The latter factor does not arise with Fehling's solution, but it becomes of importance when sodium carbonate is substituted for sodium hydroxide. Empirical tables relating to titrations at various concentrations have been prepared by Brown, Morris and Millar,² Munson and Walker,³ Pflüger,⁴ Peters,⁵ and Lane and Eynon.⁶

Various modifications of Fehling's method involving the use of potassium ferrocyanide as external indicator have been advanced. More recently Lane and Eynon have suggested the use of methylene blue as internal indicator. In a reducing medium methylene blue becomes reduced to the colourless form when it has reached a characteristic potential with respect to a platinum electrode immersed in the solution. It will be shown that the choice of methylene blue as an internal indicator was highly successful in view of the potentials indicated at the end-point of the titration of Fehling's solution with a solution of glucose.

In view of the results of some recent work of Britton and Edge⁷ on the nature of alkaline solutions of copper oxide and sodium tartrate, it appeared likely that such a titration could be followed potentiometrically. These solutions contained sodium tartrate and sodium hydroxide in varying proportions with respect to copper sulphate. The copper-ion concentrations were exceedingly small, ranging from 3.6×10^{-15} to 8.0×10^{-17} . Increasing either the relative amounts of tartrate or alkali caused the copper-ion concentrations to be somewhat depressed. Computations of the ionic product, $[\text{Cu}^{2+}][\text{OH}']^2$, indicated that these complex solutions were only just able to retain cupric hydroxide in solution, the stability of the solutions on boiling becoming less when smaller amounts of alkali tartrate

were present. The ratio of copper : tartrate : alkali in Fehling's solution represents approximately the minimum proportions of tartrate and alkali that give a comparatively stable solution on boiling.

During the progressive addition of glucose to a complex alkaline copper tartrate solution cupric ions are reduced to cuprous ions, thus $\text{Cu}^{++} \rightarrow \text{Cu}^+$. The cupric-ion concentration will be governed by the precise composition of the alkaline copper tartrate solution and the changes in the ratio of the constituents resulting from the removal of copper in the form of precipitated cuprous oxide. The cuprous-ion concentration will be regulated by the solubility product of cuprous hydroxide and the hydroxyl-ion concentration. Allmand⁸ gives 1×10^{-14} for the solubility product of cuprous hydroxide. Hence, in a decinormal solution of sodium hydroxide, which is approximately the alkali-content of the solutions used in the present investigation (see Table I), the cuprous-ion concentration would be about 10^{-18} . The potential set up at platinum electrode during the first part of the titration will be governed by the expression

$$E_{\text{Pt}} = \epsilon_{\text{Cu}^{++} \rightarrow \text{Cu}^+} - \frac{RT}{F} \log_e \frac{[\text{Cu}^+]}{[\text{Cu}^{++}]}$$

which at 90° C., the temperature at which the potentiometric titrations here described were performed, becomes

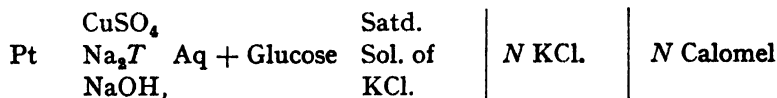
$$E_{\text{Pt}} = \epsilon_{\text{Cu}^{++} \rightarrow \text{Cu}^+} - 0.07 \log_{10} \frac{[\text{Cu}^+]}{[\text{Cu}^{++}]}$$

The end of the titration occurs when the cupric ions are completely removed, and this should be indicated by a rapid diminution in the potential recorded at the platinum electrode. After the end-point has been reached the glucose then added remains in solution, and if it enters into equilibrium with one or more of its oxidation products already formed in the solution, a potential should then be set up at the platinum electrode which will be typical of that particular equilibrium. If such a potential is very different from those prevailing during the first stage of the titration, the end-point will be marked by a steep well-defined inflexion. Preliminary experiments by Dr. H. A. Edge showed this to occur. Regarding the oxidation products of glucose, Nef⁹ was able to isolate several products by oxidising glucose with Fehling's solution.

The possibility of estimating glucose potentiometrically has been previously investigated by Daggett, Campbell and Whitman⁹ and by Niederl and Müller.¹⁰ The former workers observed that in the titration of a boiling Fehling's solution with glucose the end-point was indicated by the first large permanent deflection of the galvanometer. During the first part of the titration they reduced the small galvanometer deflections to zero by means of an adjustable resistance before each fresh addition of glucose. The latter workers describe a semi-micro potentiometric method in which a very dilute boiling Fehling's solution is titrated with glucose with the use of a platinum electrode, another platinum electrode, immersed in a solution of Fehling's solution of the same concentration, being used as the reference electrode. The inflexion indicating the end-point was not very sharp but was considered sufficiently good for accurate titration. The use of their

particular reference electrode does not permit of a study of the characteristic potentials involved.

EXPERIMENTAL.—The potentiometric titrations were carried out in a Pyrex cylindrical vessel with a capacity of about 250 ml. This was tightly fitted with an ebonite cap with holes for the insertion of one arm of the salt-bridge, a mechanical glass stirrer, two electrodes, a thermometer, a small tube connected with the burette, and with a further two holes—one for the introduction of an inert gas, *viz.* nitrogen, and the other for the removal of steam and air, and (when used) nitrogen. The cell system used for the E.M.F. measurements was



Each of the two ends of the salt-bridge, which was filled with a saturated solution of potassium chloride, was plugged with fine asbestos which had previously been immersed in a saturated potassium chloride solution. This reduced diffusion to a minimum. In order to be certain that reproducible results were being obtained two bright platinum electrodes were used and the potential difference between each electrode and the N Calomel electrode was measured after each addition of titrant. The electrodes, which consisted of platinum foil, one square centimetre in area, were attached to platinum wires sealed into the ends of glass tubes filled with mercury. The complete cell was placed in a boiling water-bath, and for the titration this was maintained at a temperature of about 90° C., and remained sufficiently constant for the purpose. During titration the burette containing the glucose solution was kept as far away from the water-bath as possible by means of a long drawn-out tube attached to the tip of the burette and leading into the titration vessel.

It was found that continuous mechanical stirring caused rather unsteady potentials to be set up at the platinum electrodes, owing to the enhanced aerial oxidation of the precipitated cuprous oxide, but that steady values were soon reached if vigorous agitation was employed during each addition of glucose and for about half a minute afterwards, and then stopped in order that steady E.M.F.'s between the electrodes and the still solutions might be attained. The E.M.F.'s were measured by means of a potentiometer.

In order to study the titration as usually performed in the presence of air, in only a few experiments was nitrogen passed into the titration vessel; 100 ml. of alkaline copper tartrate solutions were used. They were prepared from copper sulphate, sodium tartrate and sodium hydroxide in the concentrations and ratios indicated in Table I. The solution used for titration No. 2 corresponded with the molar ratio in Fehling's solution, with the exception that sodium tartrate was substituted for Rochelle salt—it having been found that the potassium present in that salt was without effect on the titration.

Typical titration curves are given in Fig. 1 (p. 23), and these refer to solutions similarly numbered in Table I. The potentials set up during the early parts of the titrations were somewhat unsteady, but both electrodes gave the same potentials

after a minute or so in still solutions. The complete reduction of the cupric oxide is marked by a well-defined inflexion extending over more than 200 millivolts, the potentials becoming correspondingly more negative. It is at this stage that cuprous oxide becomes readily oxidised by the air, so much so that the rate of oxidation may become sufficiently large to prevent the potentials from becoming constant, and they tend to fall back to smaller values. This is more marked when using the more dilute solutions of glucose. Any errors thereby introduced can, however, be minimised by adding larger volumes of glucose than are usually employed if the course of inflexion is to be minutely followed. Thus with glucose solutions of 2 g. per litre or less it is advisable to use increments of 0.2 ml. in the vicinity of the end-point. With larger concentrations the potentials are much more stable at the end-point, and additions of 0.1 ml. are permissible. It should be pointed out that when air is completely excluded by using nitrogen these difficulties are avoided.

The potentials indicated after passing the end-point do not appear to alter with increasing amounts of glucose.

Titration No. 10 is interesting in that sodium carbonate was substituted for sodium hydroxide in the complex tartrate solution. The lower pH of the solution is reflected in the lower potentials corresponding with the beginning and the end of the titration and also with the smaller range of potential over which the inflexion extends. Another significant feature is that the end-point is attained by addition of less glucose than when caustic alkali is present, for, as Table I shows (No. 10), 6.31 molecules of cupric oxide are required to oxidise one molecule of glucose compared with about 5 molecules when under the influence of free alkali (*cf.* Shaffer and Somogyi¹¹).

When once a preliminary titration has been carried out some saving of time may be effected by running into the complex alkaline solution all but about 1 ml. of the glucose solution required to complete the reaction. The course of the inflexion can then be ascertained potentiometrically.

It has also been proved by gravimetric analysis that the point of inflexion corresponds exactly with the complete reduction of the cupric oxide. Although the amount of glucose required to give the inflexion can be read directly from the titration graph, somewhat greater accuracy can be obtained by plotting $\delta(E.M.F.)/\delta x$ against x , where x is the titre of glucose, when the maximum value of $\delta(E.M.F.)/\delta x$ indicates the end-point.

Titration 1 to 9 illustrate the effects of increasing the relative amounts of (a) sodium tartrate, (b) sodium hydroxide. On comparing the titres of glucose in titrations 1, 4 and 7, it will be seen that the increasing amount of tartrate had no effect on the amounts of glucose oxidised. On the other hand, the increase in alkali concentration with each of the ratios of copper sulphate to sodium tartrate used, *viz.* 1:4, 1:10, 1:20, slightly increased the titre, or, as the last column shows, reduced slightly the number of molecules of copper oxide which oxidise one molecule of glucose. A further point, which is illustrated by the titration graphs, is that a greater proportion of sodium tartrate renders the potentials given in the early part of the titrations more negative, and this is even more marked with the greater alkali concentrations. This is to be attributed to

the greater stability of the complex copper tartrate solutions under these conditions, resulting in a smaller concentration of cupric ions.

Although alkaline solutions containing 4 to 4.6 mols. of sodium tartrate per mol. of copper sulphate, including Fehling's solution, do not readily decompose on boiling, blank experiments in which the solutions were maintained at 95° C. for the time required for potentiometric titration, *i.e.* about a half-hour, caused a little cupric oxide invariably to be precipitated. This, however, was without effect on the accuracy of the titration of these solutions with glucose. When 10 molecules of sodium tartrate to 1 of copper sulphate are used the complex tartrate solutions remain perfectly clear on boiling.

TABLE I

| No. | Concentration of alkaline tartrate solution g.-mols. per litre | | | Molar ratio $\text{CuSO}_4:\text{Na}_2\text{T}:\text{NaOH}$ | Glucose g. per litre | Amt. of glucose solution required | Mols. of CuO per 1 mol. of glucose |
|-----|--|-----------------------|------------------------------|---|----------------------|-----------------------------------|------------------------------------|
| | CuSO_4 | Na_2T | NaOH | | | | |
| 1 | 0.01 | 0.04 | 0.04 | 1 : 4 : 4 | 2 | 17.9 | 5.04 |
| 2 | 0.01 | 0.046 | 0.108 | 1 : 4.6 : 10.8 | 2 | 18.1 | 4.98 |
| 3 | 0.01 | 0.04 | 0.20 | 1 : 4 : 20 | 2 | 18.2 | 4.95 |
| 4 | 0.01 | 0.10 | 0.04 | 1 : 10 : 4 | 2 | 17.8 | 5.06 |
| 5 | 0.01 | 0.10 | 0.10 | 1 : 10 : 10 | 2 | 18.1 | 4.98 |
| 6 | 0.01 | 0.10 | 0.20 | 1 : 10 : 20 | 2 | 18.1 | 4.98 |
| 7 | 0.01 | 0.20 | 0.04 | 1 : 20 : 4 | 2 | 18.0 | 5.04 |
| 8 | 0.01 | 0.20 | 0.10 | 1 : 20 : 10 | 2 | 18.1 | 4.98 |
| 9 | 0.009 | 0.182 | 0.182 | 1 : 20 : 20 | 2 | 18.4 | 4.89 |
| 10 | 0.01 | 0.10 | 0.05 | 1 : 10 : 10 | 2 | 14.2 | 6.31 |
| | | | (Na_2CO_3) | (Na_2CO_3) | | | |
| 11 | 0.01 | 0.10 | 0.10 | 1 : 10 : 10 | 1 | 36.8 | 4.90 |
| 12 | 0.01 | 0.10 | 0.10 | 1 : 10 : 10 | 3 | 11.72 | 5.10 |
| 13 | 0.01 | 0.10 | 0.10 | 1 : 10 : 10 | 4 | 8.75 | 5.14 |

Titration 11; 5, 12, 13 (Table I) show the effect of using glucose solutions having 1, 2, 3 and 4 g. respectively per litre. As the last column shows, the amount of copper oxide required to oxidise a molecule of glucose progressively increases as more concentrated glucose solutions are used. Incidentally, these figures are in agreement with those of Lane and Eynon,⁸ using methylene blue.

Daggett, Campbell and Whitman⁹ used glucose solutions containing about 10 g. per litre. The use of concentrated solutions of this magnitude tends to give sharper inflexions, so much so that the end-point might then be considered as being produced approximately when the amount of glucose added gives the first large, permanent galvanometer deflection, as stated by them.

Potentiometric titrations were also carried out on Fehling's solution, to which methylene blue was added just before enough glucose had been introduced to produce the inflexion, in order to see whether the potential at which it becomes decolorised coincides with that of the true end-point of the reaction. The reduced form of this oxidation-reduction indicator is particularly susceptible to air-oxidation at potentials established at the end-point, as may be seen by the re-appearance of the blue colour on the surface of the liquid. When, however, the solution was kept boiling or, better, when nitrogen replaced the air in the titration vessel, the blue colour was observed to disappear at about -500 millivolts, with

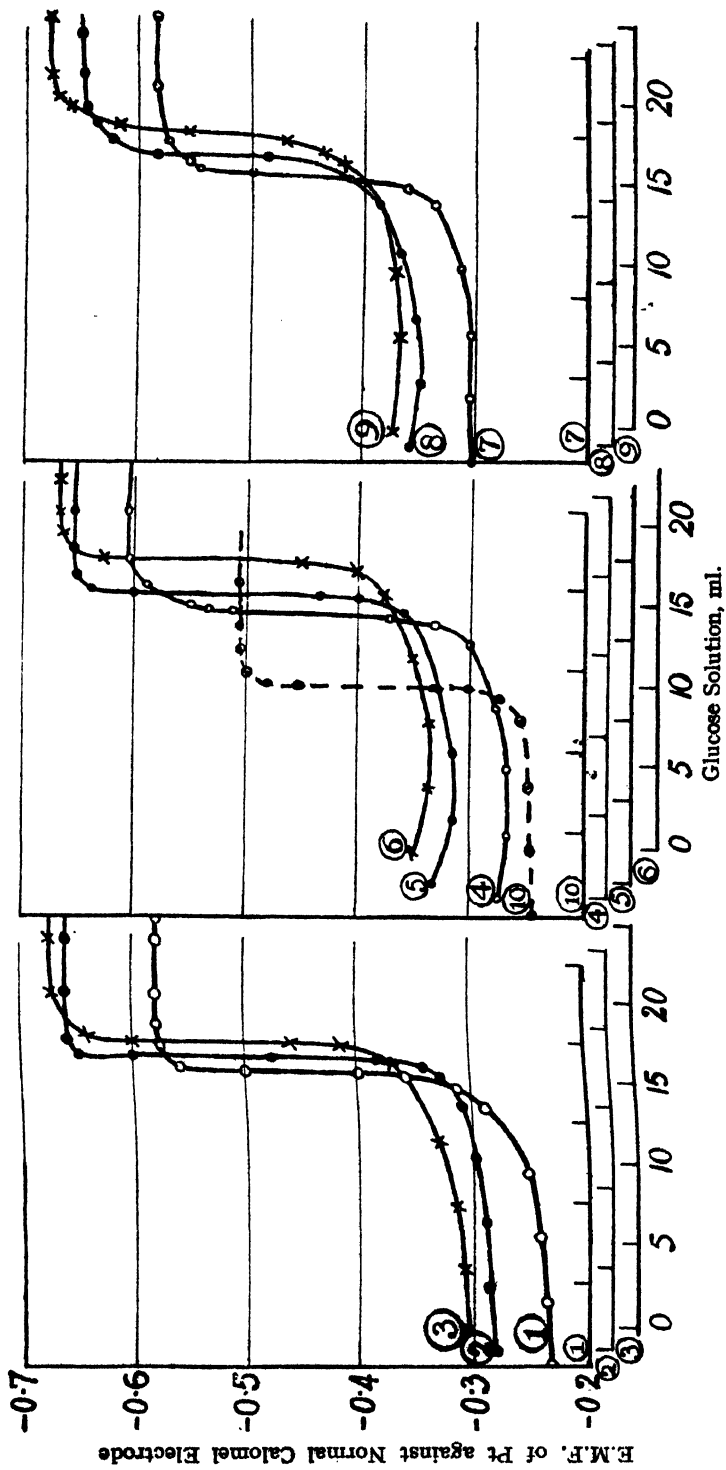


Fig. 1

respect to the *N*-calomel electrode. This is also in accordance with theory, for, according to Clark,¹² the normal reduction potential of methylene blue at *pH* 12.5 (*i.e.* the approximate *pH* of the solution) is -0.15 volt at 30°C . with respect to the normal hydrogen electrode. Temperature has little effect, giving, say, -0.17 volt at 90°C . The range of a "redox" indicator is usually restricted to 0.05 volt above and below this value, and consequently it would be expected that at 90°C . the colour of methylene blue would be completely discharged at -0.22 volt. At 90°C . the potential of the *N*-calomel electrode is 0.265 volt and therefore transition potential referred to this electrode is -0.485 volt.

In conclusion, these potentiometric titrations confirm the accuracy of the methylene blue method of Lane and Eynon for the volumetric determination of glucose. As a method of determination the potentiometric method is quite satisfactory, although it is, of course, not so rapid as that of Lane and Eynon. It is, however, essential to ascertain preferably by separate experiments, or by reference to Lane and Eynon's tables, the precise amount of cupric oxide required to oxidise glucose under comparable conditions with respect to concentration, before any calculations are made as to the amount of glucose in the unknown solution.

The ease with which cuprous oxide becomes re-oxidised by air in the vicinity of the end-point, owing to the extremely low and negative "redox" potentials then prevailing, reveal that inaccuracies must accompany the use of external indicators, *e.g.* potassium ferrocyanide.

It has further been shown that the precise ratio of sodium tartrate to copper sulphate used in the complex alkaline copper tartrate solution is not a matter of great importance, although variations in the oxidising power of the copper oxide present therein is influenced by the alkali content. The use of Rochelle salt in Fehling's solution is unnecessary, sodium tartrate being equally serviceable.

One of us (L. P.) wishes to thank the Senate of this College for a grant from the Andrew Simons Research Fund.

REFERENCES

1. H. von Fehling, *Liebig's Annalen*, 1849, **72**, 106.
2. H. T. Brown, G. H. Morris and J. H. Millar, *J. Chem. Soc.*, 1897, **71**, 281.
3. L. S. Munson and P. H. Walker, *J. Amer. Chem. Soc.*, 1906, **28**, 665; Abst., *ANALYST*, 1906, **31**, 337.
4. E. Pfüger, *Pfüger's Archiv.*, 1903, **96**, 105.
5. A. W. Peters, *J. Amer. Chem. Soc.*, 1912, **34**, 928; Abst., *ANALYST*, 1912, **37**, 277.
6. J. H. Lane and L. Eynon, *J. Soc. Chem. Ind.*, 1923, **42**, 32t.
7. H. T. S. Britton and H. A. Edge (see *e.g.* H. A. Edge, Ph.D. Thesis, London, 1938).
8. A. J. Allmand, *J. Chem. Soc.*, 1909, **95**, 2151.
9. W. L. Daggett, A. W. Campbell and J. T. Whitman, *J. Amer. Chem. Soc.*, 1923, **45**, 1043; Abst., *ANALYST*, 1923, **48**, 277.
10. J. B. Niederl and R. H. Müller, *J. Amer. Chem. Soc.*, 1929, **51**, 1356.
11. P. A. Shaffer and M. Somogyi, *J. Biol. Chem.*, 1933, **100**, 695.
12. W. M. Clark, *U.S.A. Hygien. Lab. Bull.*, 1928, **151**, 197.

WASHINGTON SINGER LABORATORIES
UNIVERSITY COLLEGE
EXETER

July, 1939

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

THE DETECTION OF UREA IN URINE STAINS ON CARPETS

IN the routine investigation of claims arising from complaints of stains appearing on carpets during use, some difficulty had frequently been experienced in satisfactorily identifying those that were suspected to have been caused by urine.

It was found from many examinations, made in comparison with known urine stains on carpets, that only a few such stains could be identified with certainty by tests depending upon their odour, appearance and deposition, reaction to ultra-violet rays, and the presence of sodium chloride. In the majority of suspected cases of urine staining, especially when the available samples were very small, i.e. one or two tufts, it was found that some of the characteristics mentioned above were not sufficiently pronounced to confirm the presence of urine.

Various tests made for urea, depending upon the evolution of ammonia, the formation of biuret, colour reactions, liberation of nitrogen, etc., whether made either directly on the stained wool tufts or on an aqueous extract from them, were entirely unsatisfactory, owing to the wool substance itself interfering with the tests. Since, however, urea is a main constituent of urine, it was considered that if a reliable method for the isolation and detection of urea from urine stains could be found it would provide a very useful test to assist in their identification.

With this in view, several experiments were made on pieces of carpets stained with human and domestic animal urine, and from the results the method here described was devised. It depends upon the extraction of urea from the stained tufts by means of alcohol and acetone and its subsequent identification under the microscope, and has proved very satisfactory in practice.

Three or four of the stained tufts are carefully removed from the carpet, transferred to a test-tube, and extracted successively three times with boiling alcohol (industrial methylated spirits), about 5 ml. being used for each extraction, and the extracts are filtered into a boiling-tube. A few pieces of porous porcelain are added, and the alcohol is carefully evaporated by heating the tube in a water-bath. Any traces of moisture left are removed from the residue by prolonging the heating, since the success of the method depends upon freedom from moisture. The dry residue is then re-extracted three times with successive small quantities (5 ml.) of pure moisture-free acetone, and the combined extracts are filtered into a boiling-tube and concentrated down to 4 or 5 ml. by immersing the tube in a warm water-bath. With the aid of a small pipette, or by careful pouring, this extract is evaporated, drop by drop, on a microscope slide heated over a water-bath, the residue obtained being confined to as small an area as possible.

On cooling, if the stains are caused by urine, there will be a pale yellow transparent ring, either complete or formed by small globules, from which urea separates in characteristic needle-shaped crystals that can be identified under the microscope.

For confirmation, the crystals are converted into the corresponding nitrate, which also has a characteristic crystalline appearance. This is done by carefully warming the crystals with a small drop of conc. nitric acid until they are dissolved. On cooling, urea nitrate crystallises out in either small separate diamond-shaped plates or a series of elongated hexagons, according to the degree of crystallisation.

A little practice is required to acquire the technique, and at first it is advisable to make comparisons with slides prepared under similar conditions from urea.

The method has been found sufficiently delicate to detect urea in the tip of a single urine-stained tuft (*i.e.* of the order of 0.1 mg.), where the quantity of urea is too small to be identified with certainty by chemical tests. If several mg. of urea, however, are isolated, some of it will be available for confirmation by the hypobromite and mercuric nitrate tests.

It was found that the double extractions with alcohol and acetone were necessary to obtain the best results. Whilst alcohol will extract urea readily and conveniently from urine-stained wool, traces of sodium chloride present in the urine, and of sodium sulphate left in the fabric from the dyeing processes, will also be extracted, especially if either the alcohol or wool is moist. These salts will then crystallise out, giving misleading results, especially in the nitrate test, in which sodium chloride is converted into sodium nitrate yielding crystals similar to those of urea nitrate. Subsequent experiments with various solvents showed that dry acetone, whilst not very satisfactory for the direct extraction of the urea from urine-stained wool, is excellent for separating urea, completely free from these salts, from an alcoholic extract of such stains.

The importance of separating urea from sodium chloride should again be emphasised, as many of the stains encountered on carpets, having the typical discoloured appearance of urine stains, are caused by common salt alone.

G. N. GEE

65, BALDWIN ROAD
KIDDERMINSTER
November 2nd, 1939

MODIFICATION OF THE STIRRING DEVICE OF THE HORTVET CRYOSCOPE

ELSDON and Stubbs (ANALYST, 1934, 54, 702) devised a mechanical stirrer and blower actuated by an electric motor, and the apparatus, although not described in detail, was stated to give excellent results.

The mechanical blowing and stirring device used in this laboratory was obtained from a British firm. Apart from its expense, it has the drawbacks of noisiness, of being suitable only for laboratories equipped with electric power, and that the high humidity conditions in the tropics are particularly favourable for the swelling of the composition movable parts of this type of blower, with consequent jamming and rupture of the rubber connections.

These drawbacks were overcome by simplification of the stirring and blowing devices. Blowing can be efficiently carried out by means of the ordinary copper water-blast and filter-pump, which is a standard fitting in most laboratories.

For the operation of a vertical-lift type of stirrer the simplest method tried was an adaptation of a vacuum-operated wind-screen wiper, such as is made by the Trico Company. This method has the advantages of being comparatively cheap and noiseless in action, and it can be operated from the suction side of the same filter and blast pump as is used for the supply of compressed air.

The fitting of the commercial motor, which is extremely simple, is briefly as follows:—The actual wiping device is disconnected and discarded, and a short arm is soldered to the motor spindle. The end of the stirrer cord is then attached to the extremity of this arm by means of a simple swivel joint. The vacuum motor is mounted on the back board of the Hortvet apparatus by means of a bracket and is connected with the suction pump by a piece of pressure tubing fitted with an adjustable clip. Complete control of the stirring rate is thus possible, a small knob, easily accessible to the operator, being used for discontinuing stirring after seeding out has taken place. Constructional details and the neatness of the device are indicated in Fig. 1.

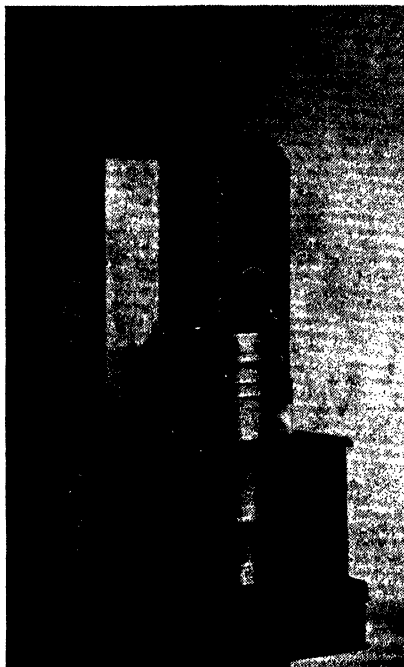


Fig. 1
Modification of the Stirring Device
of the Hortvet Cryoscope

W. J. BLACKIE
G. F. FLEMONS

CLEANING JENA GLASS FILTERS

HERBERT HAWLEY

Official Appointments

| | | | |
|---------------------------|----|----|----------------------------------|
| MIDDLESEX COUNTY | .. | .. | Miss M. H. PEARSON (Deputy P.A.) |
| BROMLEY BOROUGH | .. | .. | F. W. F. ARNAUD |
| BEXLEY BOROUGH | .. | .. | " |
| ERITH BOROUGH | .. | .. | " |
| ORPINGTON BOROUGH | .. | .. | " |
| ESHER URBAN DISTRICT | .. | .. | E. HINKS |
| " | .. | .. | D. D. MOIR (Deputy P.A.) |
| SURBITON "BOROUGH" | .. | .. | E. HINKS |
| " | .. | .. | D. D. MOIR (Deputy P.A.) |
| NEWTON-LE-WILLOWS U.D. | .. | .. | J. R. STUBBS |
| LEIGH BOROUGH (LANCS.) | .. | .. | " |
| RUGBY BOROUGH | .. | .. | W. T. RIGBY |
| " | .. | .. | F. G. D. CHALMERS (Deputy P.A.) |
| OLDBURY BOROUGH | .. | .. | H. E. MONK |
| WORTHING BOROUGH | .. | .. | S. A. WOODHEAD |
| " | .. | .. | R. F. WRIGHT (Additional P.A.) |
| ECCLES BOROUGH | .. | .. | G. H. WALKER |
| MALDEN AND COOMBE BOROUGH | .. | .. | E. HINKS |
| ILFORD BOROUGH | .. | .. | B. DYER |
| " | .. | .. | G. TAYLOR |
| KEIGHLEY BOROUGH | .. | .. | F. W. M. JAFFÉ |
| KESTEVEN (LINCS.) | .. | .. | W. W. TAYLOR (Additional P.A.) |
| LINCS. COUNTY | .. | .. | " |

Notes from the Reports of Public Analysts

The Editor would be glad to receive the Annual or other Reports of Public Analysts containing matter of special interest to the Society. Notes made from such Reports would be submitted to the Publication Committee.

COUNTY OF ABERDEEN

REPORT OF THE COUNTY ANALYST FOR THE YEAR 1938

OF the 827 samples examined during the year, 229 were purchased formally.

SULPHUR DIOXIDE IN MINCE.—As stated in previous reports, sulphur dioxide is permitted in mince during the months June to September, inclusive, to the extent of 450 parts per million. Two samples examined respectively in April and May and two in October contained from 100 to 166 p.p.m. and were condemned.

COPPER IN TOMATOES AND TOMATO PURÉE.—Eight samples of tomato purée contained from 3·60 to 5·65 p.p.m. of copper. The amounts found in foreign-grown tomatoes (fresh and preserved) were as follows:—Fresh tomatoes (2), 1·05 and 1·00; tomatoes, 0·45; Dutch tomatoes, 0·50 p.p.m.

COMPOSITION OF "SOWANS."—Oat "sids," which will keep for a long period, contain a valuable easily digestible starchy food which when prepared is known throughout Scotland as "Sowans" (cf. *The Scots Kitchen*, by F. M. McNeill, published by Blackie & Son). The solid matter in a sample of sowans gave the following analytical results:—oat fat, 2·78; mineral matter (largely phosphate), 3·43; starch, 55·83; cellulose, 37·96. The starch was in a readily digestible form.

J. F. TOCHER

CITY OF CARDIFF

ANNUAL REPORT OF THE PUBLIC ANALYST FOR 1938

LEAD IN BEER AND CIDER.—Eight samples of beer contained from 0 to 0·45 part of lead per million; two contained 4·0 and 5·4 p.p.m. and were condemned. Up to the present beer and cider containing not more than 0·05 grain of lead per gallon (0·7 p.p.m.) have not been the subject of adverse comment in Cardiff, but as the consumption of $2\frac{1}{2}$ pints of beer containing that proportion would result in the ingestion of 1 mg. of lead, it is obvious that this limit must be revised. The use of tin-washed lead pipes should be discontinued, and it is to be hoped that in the near future no lead pipes will be used in any public house in Cardiff. In the meantime the limit for lead in beverages will be reduced. It seems reasonable to adopt 0·3 p.p.m. as the limit for lead in beer.

So far as is known no cider sold in Cardiff is drawn through pipes. One sample drawn directly from a barrel contained 0·15 p.p.m. of lead.

LEAD, TIN AND ZINC IN CANNED FISH.—The amounts of lead in 12 samples of sardines and one of anchovies were all below the limit of 5 p.p.m., which has been adopted provisionally by Port Medical Officers. Four samples of anchovies and one of sardines taken from a stock condemned by the Chief Sanitary Inspector on account of the "springy" condition of the tins contained amounts of tin ranging from 12 to 18 grains per lb., while the sardines contained 8 grains per lb. These tins contained tomato sauce and all were unlacquered.

Zinc was found in amounts ranging from 17 to 62 p.p.m. in the eight samples examined. It seems probable that zinc occurs naturally in fish in these proportions.

COPPER AND ZINC IN TOMATO PULP.—At a conference of Port Medical Officers convened by the Ministry of Health it was agreed to draw the attention of importers

to the necessity for eliminating as far as possible contamination of tomato products with adventitious copper, and to require, as from January 1st, 1939, that samples taken from consignments arriving in this country should contain not more than 100 parts of copper per million parts of dry matter. It is intended to reduce this provisional limit still further.

Eleven of the 15 samples were contained in cans lacquered on the inside. The four samples in unlacquered cans contained 450, 555, 926 and 1823 parts of tin per million parts of dry solids. The contents of one of the cans that were well lacquered contained only 201 parts of tin per million of dry matter. It is possible that this was introduced during processing, but in any event the amount is very much less than the quantities present in the unlacquered cans. In one can, however, the seam had been soldered, leaving a strip of exposed metal down the inside of the can, and this was completely coated with copper, which was undoubtedly derived from the copper present in the purée.

One of the samples from unlacquered cans also contained zinc to the extent of 114 parts per million, which was equivalent to 722 parts per million parts of dry matter. The rest of the samples gave no indication of the presence of any appreciable amounts of zinc, and this proportion must be regarded as very excessive.

The concentration of these products varied enormously, the total solid matter ranging from 9.91 to 44.18 per cent. In a few instances the approximate amount of dry extract was given either on the label or impressed on the lid of the can, *e.g.* 28-30 per cent., 30-36 per cent.; in others where the labels bore the term "double extract" the dry solids ranged from 33.1 to 40.9 per cent., but with the majority of the samples there was no indication of the concentration of the pulp. It appears desirable, therefore, that makers should declare the amount of dry matter in these preparations or use recognised descriptions such as "single," "double" or "triple" extracts according to their dry tomato contents.

STANLEY DIXON

METROPOLITAN BOROUGH OF STEPNEY

ANNUAL REPORT OF THE BOROUGH ANALYST FOR THE YEAR 1938

OF the 1586 samples submitted under the Food and Drugs (Adulteration) Act, 976 were purchased formally.

QUININE AND CINNAMON TABLETS.—A formal and an informal sample from the same vendor were deficient in quinine sulphate to the extent of 32.4 and 32.8 per cent., respectively. The formula on the bottle showed 12.5 per cent. of quinine sulphate, whereas only 8.4 per cent. was found. The manufacturers stated that their declaration was based on the weight of the uncoated tablets; the uncoated tablet weighed 3 grains and sugar-coated tablets 5 grains. This is misleading, for a tablet might consist of 90 per cent. of coating. To avoid misunderstanding the label should show the actual content of quinine sulphate per tablet. The vendor was cautioned.

D. HENVILLE

Legal Notes

Under this heading will be published notes on cases in which points of special legal or chemical interest arise. The Editor would be glad to receive particulars of such cases.

BLEACH OINTMENT

A DRUGGIST was summoned at Croydon Police Court on November 8th for the sale of bleach ointment not of the nature, substance and quality demanded.

Mr. W. W. Ruff, prosecuting for the Surrey County Council, said that the amount of available chlorine in ointment prepared according to the formula in the Home Office Air Raid Precautions Handbook, No. 2, was 15 per cent. The amount in the ointment purchased from the defendant's shop was 1·6 per cent.

Mr. G. G. Baker, for the defence, pointed out that the handbook said "can be made," not "should be made." It was, in fact, put forward as a suggestion. No indication had been given to druggists in the district as to the standard the County Council was expecting to be used. In ninety-nine cases out of a hundred a prescription was made up from the British Pharmacopoeia. In this case it was made up from the formula supplied in the A.R.P. Catalogue of a reputable firm of manufacturing chemists.

The defendant said that he had never heard of the A.R.P. handbook, and had never been notified by the local authority of any standard to which he was expected to conform.

Mr. Thomas Tickle, F.I.C. (Public Analyst for the County of Devon), gave evidence that, in his opinion, the chlorine was self-destructive and disappeared, provided that the petroleum jelly had not been bleached (from yellow paraffin) by chlorine already. It made no difference whether there was 1·6 per cent. or 10 per cent. of chlorine, as with white petroleum jelly every particle of chlorine was surrounded by an oily layer.

The Bench dismissed the summons, but refused an application for costs.

At Sutton Police Court on November 8th a druggist was summoned on a similar charge. In this case the ointment supplied by the defendant contained 6·6 per cent. of chlorinated lime and 93·4 per cent. of white petroleum jelly.

The defendant said that he had no knowledge of the Air-Raid Precautions formula. He had consulted the British Pharmacopoeia and the British Pharmaceutical Codex. Neither contained a bleach ointment, but both described liquid preparations containing chlorinated lime. The greatest proportion of available chlorine in any of these formulae was 3 per cent., and he took this as a guide for a bleach ointment.

Mr. Baker, for the defence, said that the primary and essential point was clear. Here was a request for bleach ointment—not a request for bleach ointment for treating mustard gas burns or a special Home Office ointment. There was no indication that the ointment was required for a special purpose. The Bench was asked to say what was the standard formula for bleach ointment. There was no legal standard and the various air-raid precautions issued contradicted themselves.

The Chairman (Mr. Horace Sharp) said that the Bench had had very great difficulty with the case, but they had come to the conclusion that it would not be safe to convict the defendant. They felt, however, that a very useful purpose had been served by the case, for now the general public and druggists would know that something was advised in the air-raid precautions handbook which might be regarded as a standard and which it would be wise to keep to, at least until something more authoritative was found.

Milk and Nutrition

NEW EXPERIMENTS REPORTED TO THE MILK NUTRITION COMMITTEE*

THIS is the last of the reports to be issued by the Milk Nutrition Committee in connection with researches into the nutritive values of raw and pasteurised milk, planned in 1934-35. It presents the results of the complete analysis of the school feeding experiments partly reported in Part II (*cf.* ANALYST, 1938, 63, 434) and summarises the whole of the experimental work of the Committee, including results published for the first time. Finally, an attempt has been made to assess the practical importance of these researches.

The results of experiments on small animals, mainly rats, and of the chemical tests were described in Part I (*cf.* ANALYST, 1937, 62, 463), and the results of calf-feeding experiments were reported in Part III (*cf.* ANALYST, 1938, 63, 893).

CONCLUSIONS DRAWN FROM FEEDING EXPERIMENTS ON CHILDREN.—No constant differences could be detected between the growth-promoting effects of $\frac{3}{4}$ pint of pasteurised and $\frac{3}{4}$ pint of raw milk in height, weight or chest circumference. Initially, a greater proportion of girls than of boys was placed in the higher nutritional categories as assessed clinically by the doctors. The proportions in the teachers' assessments of scholastic ability at the beginning of the test were about equal for both sexes. All the supplements of milk resulted in larger numbers of children being placed in the higher nutritional and scholastic categories by the doctors and teachers than did the supplements of biscuit. The changes attributable to milk were, on the whole, greater for the $\frac{3}{4}$ pint supplements than for $\frac{1}{2}$ pint. No constant differences could be discerned between the effects, on either assessment, of $\frac{3}{4}$ pint of pasteurised and $\frac{3}{4}$ pint of raw milk. The data for the assessments of the condition of complexion, expression, posture, tonsils, teeth and eyes, as well as for "after effects of preceding illness" and "absences since last examination" were found to be ambiguous or inconclusive, or both.

The results of the present investigation, while they add nothing further to our academic knowledge of differences that may exist between raw and pasteurised milk, nevertheless demonstrate that, under purely practical conditions, there is no difference in nutritive value between them.

NUTRITIVE VALUE OF MILK FOR CHILDREN.—The great value of milk for the growth and health of growing children, already established, has been abundantly confirmed by the results of the school feeding test. This experiment, which amounted to a practical trial of the Milk-in-Schools Scheme, has shown that definite improvements in physique, in general appearance and scholastic ability, and to a somewhat less extent in muscular strength, are to be expected from the consumption of $\frac{1}{2}$ pint, or, better still, of $\frac{3}{4}$ pint of pasteurised or raw milk.

THE NUTRITIVE VALUE OF PASTEURISED AND RAW MILK.—Rapidly growing animals, such as the young calf, are particularly sensitive to food deficiencies, and for this reason the calf is the most suitable test animal that can be used to detect small nutritional differences in cow's milk. The fact that no statistically significant differences in growth rate, in general health or in the composition of the blood could be detected in groups of calves fed on raw and pasteurised milk, respectively, during a six-months' feeding trial, is therefore of great importance. It may be argued that there are defects in pasteurised milk relative to raw milk, and that these defects were neutralised by constituents of the other foods—hay and concentrates—that were fed to the calves in the latter part of the experiments.

* Part IV. The Effects of Dietary Supplements of Pasteurised and Raw Milk on the Growth and Health of School Children (Final Report): Summary of all Researches carried out by the Committee and Practical Conclusions. Published by the National Institute for Research in Dairying, Shinfield, Reading. Pp. 69, with 31 Tables. 1939. Price 2s., post free.

The answer is that it is not small defects that are important, but only those that become manifest under strictly practical conditions.

In view of these and other considerations, it is not surprising that the addition, to the house diets of school children, of $\frac{3}{4}$ pint of raw or pasteurised milk did not reveal any nutritive differences. As the supplements formed only a part of the total diet of the children, defects in one or the other would have had to be fairly gross to have influenced growth and health. The results showed that there were no differences of practical importance between them.

The matter is different for infants who might be fed exclusively, or almost so, on cow's milk. In the first place, cow's milk contains too little iron for the proper nutrition of the infant, and has therefore to be supplemented with iron in some form (*e.g.* egg yolk, as recommended by the Technical Commission of the League of Nations). Again, commercial pasteurisation, as has been reported in Part I, lowers the vitamin C content of milk, and it is usual to add some fruit juice to milk for infants to counteract this defect. On the basis of existing knowledge, cow's milk, raw or pasteurised, intended for infants, ought to receive such supplements.

The loss in vitamin C following pasteurisation is not *primarily* due to the heat treatment, but to previous exposure of the milk to light. According to present knowledge, if all the vitamin were present in the milk as ascorbic acid at the time of pasteurisation, the heating would not have any effect upon it. Under prevailing conditions, however, exposure of the milk to the shorter light rays causes some of the ascorbic acid to combine with the oxygen dissolved in the milk. The resulting oxidation product—dehydro-ascorbic acid—is easily destroyed by heat and the experimental evidence shows that it is only this fraction that is inactivated by pasteurisation. The presence of minute traces of copper (as derived from copper vessels) will also, without previous exposure of the milk to light, cause the oxidation of ascorbic acid.

The only changes attributable to pasteurisation that have been detected in these investigations are a diminution of 20 per cent. in the vitamin C content (but due primarily to exposure to light) and a slight decrease in the heat-labile fraction of vitamin B (presumably the B₁ fraction).

Department of Scientific and Industrial Research

FUEL RESEARCH

ANALYSIS OF COMMERCIAL GRADES OF COAL—PART II*

THIS Report, in continuation of the analyses published in Part I (Survey Paper, No. 37), gives the analyses of a further 277 grades of commercial coal.

The grades were sampled by the B.S.I. methods (B.S.I. Specifications, Nos. 420, 1931, and 502, 1933), and the methods of analysis used were those embodied in *Fuel Research Survey Paper*, No. 44.

Proximate analyses, total sulphur and calorific value determinations were carried out on every grade, and an ultimate analysis on "selected" grades, including both "hards" and "brights," where these occur in the same seam. The moisture-content was determined by heating the coal for 1½ hours at 105°–108° C. in a stream of oxygen-free nitrogen in the "Minimum Free Space" Oven designed at the Leeds Coal Survey Laboratory. The volatile matter in those samples obtained before May, 1937, was determined by the British Standard method B (B.S.S. No. 420, 1931), using six silica crucibles together in an electric muffle furnace at 965° C.,

* Physical and Chemical Survey of the National Coal Resources, No. 48. The Yorkshire, Nottinghamshire and Derbyshire Coalfield, Nottinghamshire and Derbyshire Area. Pp. 166. H.M. Stationery Office, York House, Kingsway, London, W.C.2. 1939. Price 3s. net.

but after that date the determinations were carried out with a platinum crucible in the muffle at 925° C. The total sulphur was determined by the Eschka method, and the forms of sulphur by that of Powell (*U.S. Bur. of Mines, Techn. Paper, No. 254, 1921*; British Standard Methods, No. 687, 1936), while the method developed at the Birmingham Coal Survey Laboratory (*Fuel Research Paper, No. 44*; Alternative Method, p. 43) was adopted for the determination of total carbon dioxide. These determinations, together with the sulphur in the ash, were required for calculating to the "coal substance" the analyses of those grades that had been subjected to an ultimate analysis.

ASH.—Variations in the ash-content are shown to depend *inter alia* on the quantity of impurity present, on the closeness of sizing, and on the method of mining. The ash, as determined by the standard method, differs from the mineral matter in the coal both in amount and nature as the result of changes (described in detail in the Report) that occur during incineration.

The fusion-point of the ash obtained from washed grades and from selected grades of large, hard steam coal may be constant within small limits, but may not be so constant in secondary grades and rough slacks, owing to variations in the amount of adventitious ash.

CHLORINE.—In the examination of seam section in the area under survey it has been found that the amount of chlorine is substantially the same throughout any one section. This regularity of the chlorine-content is sufficient to justify the use of chlorine figures determined in one grade for the calculation of data for other grades in the same seam and colliery. In South Yorkshire, also, it has been found that all grades from a single colliery contain very similar amounts of chlorine (*Fuel Research Survey Papers, Nos. 20 and 31*).

SULPHUR.—For any one seam the highest sulphur-content usually occurs in grades of second quality, which are picked out on account of the visible pyrites they contain. The smaller unwashed grades, with a relatively high ash-content, do not necessarily have a higher sulphur-content than the larger and cleaner grades.

CALORIFIC VALUE.—The calorific value of a grade "as received" will vary according to its moisture and ash contents, but the dry ash-free calorific value of any grade should be constant, within narrow limits, so long as there is no major change in the seam or in the mining conditions.

GRAY-KING CARBONISATION-ASSAY AT 600° C.—The cokes from this assay have been classified into eleven types; photographs and descriptions of these types appear at the end of the report.

New Zealand

ANNUAL REPORT OF THE DEPARTMENT OF AGRICULTURE FOR 1938-1939

THE Report of the Chemistry Section of the Department is by Mr. E. R. Grimmett, M.Sc., the Chief Agricultural Chemist; it describes various investigations carried out in association with the other sections of the Department, such as chemical work on Canterbury pastures in connection with lamb-mortality research and with animal health and nutrition.

COBALT DEFICIENCY IN PASTURES.—More than 550 pasture samples, 135 animal organs, and various soils, limonites, limestones and cobaltised superphosphates were analysed. Low cobalt figures were obtained with a number of pasture samples (below 0.08 p.p.m. of cobalt) in the spring and summer months. A further extension of the cobalt survey of North Island pastures has been made, and special attention has been given to seasonal variation in the cobalt-content of

pastures. Pasture cobalt tends to be at a minimum in late spring and early summer and at a maximum in winter or after prolonged drought.

Analyses of pastures from cobaltised superphosphate top-dressing experiments have shown a high initial uptake of cobalt, but later a rapid fall especially during the period of flush growth. The experiments themselves have been outstandingly successful, and have afforded further evidence of the efficacy and practicability of this method of preventing bush sickness.

Cobalt in New Zealand Soils and Rocks.—Analyses of several New Zealand ultra-basic magnesian rocks gave figures ranging from 67 to 115 p.p.m. of cobalt. Some North Auckland lateritic soils associated with cobalt-deficient pastures have shown figures as high as 10 p.p.m. for total cobalt, but the cobalt present is relatively insoluble, even in strong acid. Owing to the occurrence of cobalt deficiency in the Wairarapa on a paddock built up of a fine limestone wash, analyses of typical limestones were made. Some low figures were obtained, the amounts ranging from 0.2 to 5 p.p.m. of cobalt.

Cobalt in Animal Organs.—The average cobalt-content of the livers of bush-sick sheep is about 0.04 p.p.m. or less, but for healthy animals more than 0.10 p.p.m. A similar difference has been observed for cattle. There is no evidence of copper deficiency associated with bush sickness in the North Island.

FACIAL ECZEMA.—During an extensive outbreak of facial eczema in the Waikato in April large samples of pasture were obtained from fields believed to be still active in causing the disease. Analyses showed that icterogenin (the substance which, it was suspected, might be the active factor causing liver derangement) was not present in detectable amounts. It is possible, however, that the pasture might have passed the active stage when the samples were collected; that an icterogenic substance other than icterogen itself might have been present; or that a toxin may be formed after the pasture has been eaten by the animal. Analytical work in association with records of changes in animal health (if any), changes in botanical composition, and changes in growth rate, succulence, etc., is in progress.

VARIATION IN SOIL NITRATE.—Concurrently with the pasture analyses a series of determinations of soil moisture, nitrate and ammonia has been made in some of the experimental paddocks. A feature that has emerged is the extreme variation in soil nitrate found at intervals of a few hours throughout a single day. This may change from a level as low as 10 p.p.m. in the early afternoon to one of 80 p.p.m. two hours later.

STANDARDS FOR TRACE ELEMENTS IN PASTURES.—In co-operation with the Cawthron Institute and the Dominion Laboratory, work has been begun to obtain quantitative data on the occurrence of a number of elements that are present in many pastures and in animal tissues, but of the possible function of which little is known. Naturally healthy and productive pastures have been selected in several localities, and by the use of movable enclosures clean samples are being obtained several times a year. Determinations of cobalt and of some other elements are being made by chemical methods, and the samples are then handed over to the Dominion Laboratory for spectrographic analysis of the ash.

ARSENIC IN SOILS AND WATERS.—The investigation into the alleged poisoning of live-stock by arsenic occurring naturally in soils and waters has been continued, and a comprehensive report has been submitted to the Lands Department. This includes a map and tables showing the distribution of arsenic in soils, muds and waters; ante- and post-mortem notes and figures for arsenic in the organs of 31 cases of live-stock mortality; the arsenic-content of pasture samples and accounts of experiments on cattle at Reporoa and Wallaceville. Sixty-five samples of muds and soils were collected. The arsenic in muds varied from 0.0068 per cent. to 1.9 per cent. Representative samples of the whole soil to a depth of 9 inches in pasture lands gave results for arsenic ranging from 0.0008 to 0.525 per cent. Thirty-nine samples of water from springs, streams and surface depressions showed

amounts varying from a trace to 2.6 grains of As_2O_3 per gallon. Arsenic was present in the mud deposits in several places, either in combination with iron in impure limonite (the arsenic readily soluble in dilute acid) or as a sulphide (insoluble in dilute acid). In some places amounts of orpiment and realgar up to 5 per cent. were present in orange-yellow siliceous deposits.

The general finding was that arsenic is of widespread occurrence in the spring and drainage waters and the soils of the lower-lying portions of the Reporoa Settlement, but that it is responsible for only a small annual mortality, together with further, but undefined, unthriftiness in live-stock. Clean-growing pasture on arsenical soil normally contains too little arsenic to have any toxic effect, and crops, such as oats, from such soils are also harmless. Farmers are recommended to fence off areas where arsenical springs occur, or where the mud along stream edges or in depressions is strongly arsenical, to provide safe water, and not to graze recently flooded areas while the pastures are still muddy.

DETERMINATION OF AVAILABLE PHOSPHATE.—In the soils sent in for examination the usual trouble was phosphate deficiency. Egner's lactate method for estimating available phosphate has been studied and found to show considerable promise. Certain South Island soils showed a very high phosphate-content when extracted with 1 per cent. citric acid solution, but a deficiency by the lactate method. It has not yet been ascertained which is the truer test.

BROMOTHYMOL BLUE TEST FOR MASTITIS.—The staff of the Veterinary Laboratory has been studying the anomalies shown in the bromothymol blue test for mastitis. Frequently the test does not pick out an obviously infected animal. Work done in conjunction with the Dairy Research Institute showed that only about 50 per cent. of the results were correct when checked by the leucocyte assessment in the diagnosis of mastitis. The check results obtained in three other laboratories showed still poorer correlation figures. The factors that made colour testing inaccurate were (a) the fact that the milk in many cases of mastitis does not have an alkaline reaction; (b) the reading is upset by the quantity of cream, by the interval of time between the taking of the samples and the reading, by the quantity of milk and bromothymol blue taken; (c) by the personal factor, many officers being unable to distinguish shades of colour.

CHEMICAL CONTROL OF RAGWORT.—A farm experiment confirmed the superiority of sodium chlorate as compared with dichromate, thiocyanate, and bisulphite weed-killers. Another experiment is in progress to determine the effect of treatments of individual plants at various times of the year and the toxic dose per plant. Experiments in the Ruakura nursery indicate that injections of chlorate into the soil or applications on the soil surrounding the plants are not as effective as those applied to the foliage in the usual way, that dilution of the spray has little effect, and that the seed from flowering plants sprayed with 2.5 and 5 per cent. sodium chlorate solutions has a germination of 10 to 20 per cent., as compared with 50 to 80 per cent. in unsprayed seed-heads.

Ragwort Feeding to Cattle and Sheep.—A cow that was kept on a diet of 1 lb. of rosette-stage ragwort per day and afterwards given ragwort infusion over a period of three months, has recently calved and appears to be quite normal. Two sheep were kept on ragwort for $2\frac{1}{2}$ years and were then slaughtered and their livers examined. One sheep was normal, but the second showed some increase in fibrous connective tissue. Apparently the amount of toxic substance in rosette-stage ragwort is very small, and insufficient to cause any serious damage to sheep or cattle unless fed in large quantities.

Government of Madras

ANNUAL REPORT OF THE CHEMICAL EXAMINER FOR THE YEAR 1938

THE Chemical Examiner (Mr. S. Rajagopal Naidu, F.I.C.) reports an increase in the work of the department, the total number of cases investigated being 1785, as compared with 1670 cases in 1937.

HUMAN POISONING CASES.—Poison was detected in 241 of the 467 cases investigated. *Datura* heads the list with 36 cases; then come oleander with 30, opium with 29, and madar juice with 14 cases. Poisoning with organic poisons was nearly twice as frequent as with inorganic poisons. Among the latter, mercury claimed 21 cases, arsenic 20, copper sulphate 12, cyanide 10, and nitrite 9 cases.

***Datura* Poisoning.**—A study of *datura* cases has shown that the type of criminal who uses *datura* generally carries with him the powdered seeds, and probably buys sweets at some bazaar to mix with the powder. If only a portion of the powder has been used, the remainder may be expected to be found in his possession. In a case in 1937 the culprit, caught immediately after the offence, had on him a packet of the powdered seeds. In a 1938 case a boy was given sweets by a religious mendicant and soon afterwards became giddy and then unconscious. He was removed to hospital, where, after treatment, he recovered. A mydriatic alkaloid was found in his urine, and particles of *datura* seed were detected in the stomach washings. Some villagers chased the culprit and found in his possession a packet of vegetable powder.

***Oduvan* Poisoning.**—There were three cases of poisoning by *oduvan* leaf (*Cleisanthus collinus* Benth.), all of them suicidal. The reactions of *oduvan* were obtained with the viscera, and particles of leaf with the microscopical structure of *oduvan* were isolated from each of the stomach contents.

The method employed for studying the poisonous extract from *oduvan* leaf was the same as that used for the extraction of "nerin" from *Nerium odorum* (cf. ANALYST, 1939, 64, 121). A yellowish-white crystalline substance (m.p. 192–194° C. with decomposition) was obtained. It was freely soluble in alcohol and chloroform, but sparingly soluble in water. It did not reduce Fehling's solution until hydrolysed with emulsin or hydrochloric acid. Its elementary composition was: C, 53.9; H, 6.91; O, 39.19 per cent., corresponding with the tentative formula $C_{22}H_{34}O_{12}$. The name "oduvin" is suggested for this substance. Titration, by Lane and Eynon's method, of the hydrolysed product gave one mol. of an aldohexose calculated as dextrose, from one mol. of the original substance.

In addition to its colour reaction with sulphuric acid, described in the 1937 report (ANALYST, 1939, 64, 121), *oduvin* gives a bright vermilion colour with fuming nitric acid or with a mixture of equal parts of conc. sulphuric and nitric acids.

***Melia Composita* Poisoning.**—A man drank the juice of the bark of "malai vembu" (*Melia composita* Willd.) and died in about an hour and a half. No poison was detected in the viscera, but from the vomit a crystalline poisonous substance was extracted resembling the product obtained by extracting the bark with acid ether. Both extracts produced in frogs convulsions similar to those caused by picrotoxin, but whilst picrotoxin gives a red colour when treated with a dilute solution of benzaldehyde and a drop of conc. sulphuric acid (Melzer's test), the two extracts gave a permanganate purple colour. Moreover, the extracts did not give any colour on treatment with potassium nitrate and sulphuric acid and then with excess of potassium hydroxide, whereas picrotoxin gives a brick-red colour in this test. The extract from the bark did not reduce Fehling's solution either before or after hydrolysis.

Poisoning by *Manihot utilisima*.—The "jungle potato" is used extensively as a food in Malabar and Cochin, and, according to local information, there are two

varieties, one of which is poisonous. There is no appreciable difference between the two, except that the poisonous variety is darker, has redder leaves, and is not so soft when cooked as the non-poisonous variety. According to the local medical officer there have been instances of goats dying after eating the leaves of the poisonous variety, but there is no record of human beings having been poisoned after eating the root when cooked.

A woman and two children were stated to have died after eating uncooked "jungle potato." The bodies, when submitted, were in an advanced state of decomposition. No cyanide or other poison could be detected in the viscera. The root suspected to have caused death was identified as *Manihot utilisima*. When crushed, acidified with phosphoric acid, and distilled it yielded hydrogen cyanide in an amount corresponding with about 14 parts per 100,000 of the root. The poisonous principle thus appears to be a cyanogenetic glycoside. About a pound of the root eaten in the raw state would probably be dangerous to human life.

Bamboo Shoots.—The poisonous crystalline substance previously described (cf. ANALYST, 1937, 62, 742) has been further studied. It melts at 115–116° C., and has a composition corresponding with the formula $C_8H_{11}O_5$. The name "bambusin" is suggested for it. The red colour that it gives with strong sulphuric acid and manganese dioxide can be brought out more distinctly by substituting ceric oxide for manganese dioxide. The substance does not reduce Fehling's solution either before or after hydrolysis.

THE OLEANDERS.—The term oleander is applied to three different plants: (i) red-flowered oleander (*Nerium odorum*), (ii) odallam (*Cerbera odallam*) and (iii) yellow-flowered oleander or yellow oleander (*Cerbera thevetia*).

(i) A note on red-flowered oleander (*Nerium odorum*) and its poisonous principle "nerin" was published in the Annual Report for 1937 (cf. ANALYST, 1939, 64, 121).

(ii) *Odallam* (*Cerbera odallam*) is a tree with fleshy lanceolate leaves, large white flowers and green fibrous fruits enclosing a kernel. The kernel of the fruit is used as poison. The kernel on extraction with petroleum spirit or ordinary ether gave a non-poisonous oil with the following constants:

Constants of Oil from *Cerbera odallam* Kernels.—Iodine value (Rosenmund and Kuhnhehn), 29.1; saponification value, 178.6; Reichert–Meissl value, 1.21; Polenske value, 1.65; n_D^{25} , 1.4660; unsaponifiable matter, 0.73 per cent. (iodine value, 21.8).

An alcoholic extract of the de-fatted kernels gives, on solution in absolute alcohol and treatment with anhydrous ether, an abundant white precipitate which requires for purification only a few precipitations from absolute alcohol with anhydrous ether and is free from nitrogen.

This extract is freely soluble in alcohol and water, but insoluble in ether, benzene or chloroform. The elementary composition and approximate molecular weight (determined by the cryoscopic method with water as solvent) suggests the molecular formula $C_{20}H_{38}O_{14}$. The name "cerberin" is tentatively suggested for this substance.

"Cerberin" reduces Fehling's solution only after hydrolysis with emulsin or with hydrochloric acid. Hydrolysis with emulsin gives colourless products, whereas hydrolysis with hydrochloric acid gives a blue colour and then a dark precipitate. The solution of "cerberin" after hydrolysis with emulsin does not give a blue colour with hydrochloric acid and is no longer poisonous. It appears to be similarly affected by the digestive secretions of animals. The lethal dose for frogs weighing about 10 g. is about 0.025 mg. Paralytic symptoms in dogs were not so pronounced as with "nerin" or "thevetin."

(iii) **Yellow-flowered Oleander (*Cerbera thevetia*).**—The kernel of the fruit is the part most commonly used as poison, though in a few cases the leaves, bark and root have also been used. Cases of accidental poisoning of children through eating the flowers are also known. Two poisonous extracts have been obtained from the

kernels: *Extract A*, from the pressed kernels, by extraction with a mixture of ether and chloroform and removal of the traces of oil by petroleum spirit, and *Extract B*, a precipitate obtained by adding anhydrous ether to a solution in absolute alcohol of the alcoholic extract of the residue from the previous extraction. *Extract A* appears to be identical with thevetin described by Dragendorff, but the molecular weight, as determined by the method of Pregl and Rast (in which camphor is used as the solvent), suggests that the formula should be $C_{27}H_{42}O_{12}$ instead of $C_{54}H_{84}O_{24}$ as proposed by Dragendorff. Thevetin is sparingly soluble in water but soluble in ether and readily soluble in chloroform. After hydrolysis with hydrochloric acid or emulsin it reduces Fehling's solution. The sugar residue after hydrolysis gives with thymol and sulphuric acid the reaction given by aldohexoses. About 0.02 mg. is fatal to frogs weighing about 10 g. Ten mg. injected into a dog weighing about three kg. produced defecation, vomiting with retching, frothy salivation, paresis of the hind limbs and terminal spasms. Death occurred in about ninety minutes. Post-mortem examination revealed engorged heart stopped in diastole, intensely congested lungs, congested liver with the gall bladder full, a pale stomach and intestines and somewhat congested kidneys. Thevetin, therefore, appears to be somewhat similar to "nerin" in its poisonous character.

The following colour reactions serve to distinguish thevetin from "nerin":

| Reagent | "Nerin" | Thevetin |
|----------------------|--|---|
| Conc. sulphuric acid | Immediate crimson. | Yellowish-brown slowly changing to mauve. |
| Keller's reagent | Immediate crimson in sulphuric acid layer and slow green in acetic acid layer. | Immediate blue in acetic acid layer and slow mauve in sulphuric acid layer. |

Extract B was purified by re-dissolving in absolute alcohol and re-precipitating with anhydrous ether a number of times. This extract is soluble in water and alcohol but insoluble in ether, benzene and chloroform. The extract thus purified was free from nitrogen and gave C, 48.6; H, 7.73; O (by diff.), 43.7 per cent. This substance on hydrolysis with emulsin gave an aldohexose, and titration of the sugar by Lane and Eynon's method (the hexose being calculated as dextrose) suggests the tentative molecular formula $C_{20}H_{38}O_{14}$. This substance also agrees with "cerberin," obtained from *Cerbera odallam*, in giving a blue colour with hydrochloric acid and in its toxicity. These facts suggest that *Extract B* of yellow oleander kernels is identical with "cerberin" obtained from *Cerbera odallam*.

Yellow oleander kernels yield (1) thevetin and (2) "cerberin," whereas *odallam* kernels yield "cerberin" but not thevetin.

EXAMINATION OF HAIR.—A 10 per cent. aqueous solution of potassium cyanide has been found a good clearing agent for the examination of dark specimens of hair. Immersion for about an hour in a solution of one drop of 20 per cent. hydrogen peroxide and three drops of dilute ammonia solution in three ml. of water is also useful for clearing the hair without affecting its structure. For obtaining cross-sections of hair good results are obtained by embedding the sections in a mixture of equal parts of rosin and hard paraffin wax, melted together. The paraffin-rosin sections may be mounted without further treatment in Canada balsam, as the mixture, being transparent, does not interfere with the microscopical study of the hair.

ULTRA-VIOLET LIGHT AND PRINTING INK.—An official minutes book was submitted to ascertain if the sheet of paper bearing pages numbered 11 and 12 was different from the other sheets. On examination under ultra-violet light pages 11 and 12 were found to be, in fact, pages 111 and 112, the extreme left figure of each having been erased. Though the carbon had been removed, the oil of the printing

ink had soaked through the paper and showed under ultra-violet light an unmistakable fluorescence corresponding with the erased figure. There was also evidence of the original paper corresponding with pages 11 and 12 having been torn out and replaced by the present paper, which was similar in other respects, by pasting at the extreme left edge.

British Leather Manufacturers' Research Association

DECONTAMINATION OF LEATHER CONTAMINATED WITH MUSTARD GAS*

TRIALS have been made by the Association in conjunction with the Chemical Defence Research Department, Technical Advisers to the Home Office, and the results are embodied in the present Report.

Decontamination from mustard gas by immersion in boiling water cannot be applied to leather articles because this treatment damages the leather. Although solutions of certain chemicals which destroy blister gas are effective decontaminants of leather, they must be removed by subsequent treatment. Since, however, warm water is equally effective a method involving its use has been developed. At 50° C. (120° F.)—a temperature that can be applied to most types of leather without causing damage—decontamination of the leather from mustard gas is complete within six hours. In the process described the leather articles are first soaked in cold water for one hour and then kept for six hours in water maintained at 50° C. A suitable plant for the purpose is described.

The process effects satisfactory decontamination of leather articles contaminated, even heavily, with drops and smears of mustard gas, but cannot be successfully applied to grossly contaminated articles, which have absorbed large amounts of liquid gas. All types of leather can be satisfactorily treated, except white kid (alum tanned), which becomes shrunken and brittle, and some chamois leathers, which may shrink. Damage may also occur to made-up leather articles that contain components (such as glue or cardboard) which are not water-resistant. The appearance and flexibility of treated articles may be improved by cleaning, and polishing or oiling after drying.

Water dripping from the soaked articles may be contaminated and adequate precautions should be taken to avoid spread of contamination from this source.

Food and Drugs Act, 1938

APPLICATION TO SCOTLAND

THE following provisions of the Act became applicable to Scotland on October 1st, 1939:

Section 8.—Power of the Department of Health for Scotland to make regulations as to the importation, preparation, storage, sale, delivery, etc., of food and for prohibiting or restricting the addition of any substance to, and regulating generally the composition of, any food (other than bread or flour).

Section 30.—Power of the Department of Health for Scotland to make regulations as to the composition of bread and the addition of substances to flour.

Section 31.—Prohibition of adulterants in bakehouses and mills.

Section 33.—Conditions to be observed in dealings in margarine, margarine cheese and milk-blended butter.

* British Leather Manufacturers' Research Association, 1-6, Nelson Square, London, S.E.1.

Part IV.—Provisions as to Importation.

Section 81 (4).—Presumption as to sale for human consumption.

Section 92 (1) to (4).—Supplementary provisions applicable to Food Regulations and Bread and Flour Regulations.

Section 98.—Repeal of two Sections of the Bread Act, 1836.

Section 100 (1).—Definitions of "container," "cream," "preparation" and "substance."

Section 100 (2).—"Milk" refers also to cream and separated milk, but not to dried milk or condensed milk. "Food" includes food "for the manufacture of products for human consumption."

Section 101.—Repeals extending to Scotland—The Bread Act, 1836; The Public Health (Regulations as to Food) Act, 1907, in so far as it empowers regulations to be made regarding food (excluding milk); The Bread Acts (Amendment) Act, 1922; part of The Food and Drugs (Adulteration) Act, 1928, *viz.* Section 6 (except subsections (2) and (7)), Sections 12, 20 and 23, the proviso to subsection (3) of Section 27, and subsections (5) and (6) of Section 28.

Section 103 (1).—Name of the Act and date of operation.

These provisions (other than Part IV) are to be construed as one with the Food and Drugs (Adulteration) Act, 1928, and shall be deemed to be included in Part I thereof, subject to certain modifications detailed in Section 102.

APPLICATION TO NORTHERN IRELAND

The Act does not apply to Northern Ireland except as regards Part IV (Provisions as to Importation), which is slightly modified as detailed in Section 102. Part of The Food and Drugs (Adulteration) Act, 1928, is repealed, *viz.* Sections 12 and 20, the proviso to subsection (3) of Section 27, subsections (5) and (6) of Section 28, and Section 36.

National Institute for Medical Research

INTERNATIONAL STANDARDS FOR PROLACTIN AND FOR THE GONADOTROPHIC SUBSTANCE OF PREGNANT MARES' SERUM

It was stated in a previous issue (*ANALYST*, 1939, 64, 430) that the Third International Conference on the Standardisation of Hormones, held at Geneva in 1938, had decided that international standards should be established for certain hormones of the anterior lobe of the pituitary gland and analogous substances found in urine and serum, and that international units should be defined in terms of a weight of each such standard. It was further decided that the final preparation of these standards, their dispensing in a form suitable for the use of the laboratory worker, their storage, preservation and subsequent distribution should be undertaken by the National Institute for Medical Research, Hampstead, London.

The first of these new standards, as already announced, *viz.* that for the gonadotrophic substance of human urine of pregnancy—chorionic gonadotrophin—was established in May of last year.

We are now asked to announce that the preparation of two additional international standards has been completed, *viz.* for the gonadotrophic substance of pregnant mares' serum and for the lactogenic (crop-gland stimulating) substance of the anterior lobe of the pituitary gland. The former standard has been prepared from substantial amounts of material generously provided by five manufacturing firms in four different countries, and the latter from material supplied by seven manufacturing firms and two research institutes in five countries. The individual samples for each standard were examined by members of the Conference, and a

suitable mixtures were then made to serve as the respective international standards and finally dispensed in the form of tablets which have been packed in sealed tubes. For each standard each tablet contains approximately 100 international units.

The international standard for the gonadotrophic substance of pregnant mares' serum is dispensed in the form of 25-mg. tablets, each sealed tube containing ten of the tablets, and the international unit has been defined as the specific gonadotrophic activity contained in 0.25 mg. of the standard preparation.

The international standard for prolactin is dispensed in the form of 10-mg. tablets, each sealed tube containing ten of the tablets, and the international unit has been defined as the specific activity contained in 0.1 mg. of the standard preparation.

As with the international standards for other hormones, drugs and vitamins, the above international standards are held, on behalf of the Health Organisation of the League of Nations, at the National Institute for Medical Research, Hampstead, London, N.W.3, and are distributed therefrom to national control centres established in other countries for local distribution to laboratories, institutes and research workers; also to workers in other countries in which the establishment of national control centres has not yet been completed.

With regard to the supply of these new standards to those requiring them in the United Kingdom, applications should be made to the Department of Biological Standards, the National Institute for Medical Research, Hampstead, London, N.W.3.

British Standards Institution

The following British Standard Specification has been issued:*

No. 875—1939. STANDARD DIMENSIONS FOR SILICA BASINS, CRUCIBLES AND CAPSULES.

The series for the basins is based on specified relations of dimensions. Five sizes are provided, viz. 20 ml., 50 ml., 75 ml., 100 ml. and 200 ml.

Two types of crucible—tall and squat—are provided, with dimensions based respectively on the following ratios: $\frac{\text{External diameter top}}{\text{Overall height}} = \frac{1}{1}$ and $\frac{5}{3}$ respectively. Five sizes of tall crucibles, viz. 10 ml., 15 ml., 20 ml., 30 ml. and 50 ml., and four sizes of squat crucibles, viz. 10 ml., 15 ml., 25 ml. and 40 ml. are provided. Both basins and crucibles are made of transparent or translucent silica.

Capsules with lid for combined moisture and ash determination are made of translucent silica unless otherwise specified. They are provided in four sizes of specified dimensions.

* Publications Department, 28, Victoria Street, London, S.W.1. Price 2s.; post free 2s. 2d.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Sulphur Content of Foods. M. Masters and R. A. McCance. (*Biochem. J.*, 1939, 33, 1304-1312.)—Sulphur has been determined in over 300 different varieties of foods. In most instances a hydrogenation method was employed, whilst in the others destructive oxidation of the organic matter with a mixture of nitric and perchloric acids was used, followed by gravimetric estimation of the resulting sulphates (*cf.* Masters, *Biochem. J.*, 1939, 33, 1313). Drying prior to the analysis of some foods introduced errors into the determination, owing to the presence of volatile or potentially volatile sulphur compounds. Fruits, nuts, cereals, meat and beer lost no sulphur on drying, whilst fish and many vegetables lost small quantities, and onions, horse-radish and mustard and cress lost large amounts. The nitrogen:sulphur ratio was found to be remarkably constant for meat (15.3:1) and for fish (13.8:1), but it varied so widely with material of plant origin as to make any generalisation impossible. For animal tissues, raw or cooked, an estimate of the sulphur-content can usually be made with sufficient accuracy by multiplying the nitrogen-content by the appropriate factor. Some of the most important results are summarised below, the sulphur-contents being expressed in mg. per 100 g.:—

Fruits, raw.—Apples, cherries, damsons, grapes, grapefruit, melons, oranges, peaches, pears, pineapples, plums, rhubarb, 3.7 to 9.1; bananas, blackberries, cranberries, figs, gooseberries, lemons, loganberries, raspberries, strawberries, tomatoes, 10.7 to 18.1; currants (black, red and white), 23.6 to 33.1. *Fruits, dried.*—Prunes, 18.5; currants, raisins, sultanas, dates and figs, 23.0 to 80.8; apricots (probably sulphited), 164.0; peaches (probably sulphited), 240.0. *Tinned fruit.*—Apricots, pineapple, loganberries, peaches, pears, 1.0 to 3.0. *Nuts.*—Chestnuts, cobs, coconuts, almonds, Barcelonas, Brazils, peanuts and walnuts, 104.0 to 377.0. *Vegetables.*—Artichokes (boiled), French beans (boiled), broad beans (boiled), runner beans (raw or boiled), beetroot (boiled), cabbage (boiled), carrots (raw and boiled), cauliflower (boiled), celery (raw and boiled), cucumber, endive, lettuce, onions (boiled), parsnips, potatoes (boiled), turnips (boiled), 5.0 to 30.4; asparagus (boiled), brussels sprouts (boiled), lentils (boiled), mushrooms (fried), onions (fried), spring onions, peas (boiled), radishes, spinach, 37.5 to 87.8; watercress, 127.0; mustard and cress, 170.0; horseradish, 212.0. *Cereal and starch products.*—Biscuits, 31.8 to 107.0; bread, 54.5 to 114.5; flour, 108 to 123. *Dairy products.*—Butter, 9.1; cheese, 177 to 321; egg yolk, 164.5; egg white, 182.5; milk, 29.2. *Meats.*—Beef, 203 to 341; mutton, 164 to 197; pork, ham, 195 to 233. *Fish.*—Cod, haddock, halibut, hake, herring, kippers, mackerel, plaice, salmon, trout, 169 to 256; shellfish, 265 to 401.

F. A. R.

Gases in the Commercial Handling of Citrus Juices. G. N. Pulley and H. W. von Loesecke. (*Ind. Eng. Chem.*, 1939, 31, 1275-1278.)—The gases present in Florida grapefruit and orange juices extracted and de-aerated by

various methods have been studied. The apparatus used for determining the gas-content of the juices was a modified Peterson equipment with a Sprengel pump attached. Commercially extracted juices were found to contain (a) before de-aeration, and (b) after de-aeration: total gas, (a) 28.5 to 42.8, (b) 4.0 to 18.2; carbon dioxide, (a) 14.7 to 28.2, (b) 3.0 to 8.1; oxygen, (a) 2.46 to 4.17, (b) 0.09 to 2.39; nitrogen, (a) 9.7 to 11.3, (b) 0.9 to 7.8 ml. per litre, at 0° C. and 760 mm. The plate type de-aerators were found to be greatly overloaded in practice, and centrifugal de-aeration was the most effective type in use, the optimum conditions for which required a temperature of the juice of not less than 16° C., with a vacuum of at least 635 mm. The plate type and perforated cylinder type of de-aerator were about equal in efficiency. Oxygen in canned juices rapidly disappears, and the rate of disappearance is more rapid at higher temperatures and may adversely affect the juice, particularly as regards its vitamin C content. D. G. H.

Hydrogen "Swells" in Canned Fruits. T. P. Hoar. (*Tin and its Uses*, 1939, [3], 7.)—Representative samples of pack-rolled tinplate obtained from various sources were made into cans of a common type and packed with various fruits at Campden Research Station. These cans were stored under tropical conditions (95° F.), and as each can became a "swell" it was removed and examined. More than 1100 chemical analyses and nearly 1000 corrosion tests were made, and the separate influence of each variable was worked out by statistical methods. The main conclusions are as follows:—(1) The comparative susceptibility of a can to hydrogen swell may be assessed roughly by a corrosion test of the steel-base with a pure citric acid solution; (2) Steel-base with high copper (up to 0.2 per cent.) and also low phosphorus (less than 0.045 per cent.) gave the slowest rate of hydrogen swell in lacquered-can packs of blackcurrants, gooseberries, loganberries, Pershore plums, raspberries and strawberries; (3) Sulphur-content within the limits investigated (0.02 to 0.09 per cent.) was without influence; (4) The pack of white cherries was not influenced by the copper, phosphorus or sulphur content of the steel-base. The investigation showed that the use of tinplate with a relatively high copper and low phosphorus content will, on the average, roughly double the usual life of such packs.

Natural Ageing of Wines. E. K. Nelson and D. H. Wheeler. (*Ind. Eng. Chem.*, 1939, 31, 1279–1281.)—The chemical changes that take place during the ageing of wines in vats have been studied. Samples of red and white wines from New York and California were analysed every 3 months during the first year, and at intervals of 6 months during the second year of ageing. Oxidation-reduction ("redox") potential measurements failed to reveal any detectable amount of a reversible redox system at the pH of the wines. Sulphuring caused a temporary increase in volatile acids, and, if tannin was used in clarifying, in an increase in colour and tannin. A reduction in sugar-content was noted in the Californian wines, but the sugar did not entirely disappear in 2 years. One sample showed an aldehyde content of 0.005 g. per 100 ml. after 6 months' ageing, the previous sample having only 0.0005 g. and the subsequent one a very slight trace, possibly owing to heating in transit. The aldehyde figures for another sample were very erratic. The wines from California had a considerably smaller percentage

of tartaric acid than those from New York. Except in two samples a steady increase in the amount of volatile esters was observed during ageing. Decreases in extract, alkalinity of the water-soluble ash, total tartaric acid, potassium acid tartrate, colour and tannin occurred during ageing, as the result of oxidation and precipitation of wine lees, which consisted of potassium acid tartrate and oxidised colour and tannin. Wines in hermetically-sealed glass containers, kept at room temperature, showed a greater increase in volatile esters than those stored in vats; this is in keeping with the improvement in quality found to occur in bottled goods during storage.

D. G. H.

Rice Bran as a Raw Material of Oil. T. Hidaka. (*J. Soc. Chem. Ind. Japan*, 1939, 42, 237-239B; cf. ANALYST, 1939, 64, 750.)—The free fatty acids in the oil of rice bran kept in gunny bags increases rapidly on storage at a far quicker rate than in oil from meal treated with castor seed lipase and kept under similar conditions, but if water is added to each and the paste kept, the opposite results are noted. The decomposition power of the castor bean lipase is, in fact, far stronger than that of rice bran lipase. If the rice lipase is destroyed by heating, the increase in acidity in the oil is very greatly diminished. If fresh rice bran from the mill is pressed at once or after destruction of the lipase, a good oil is obtained with only a small percentage of free fatty acids, and this can be purified to produce a good edible oil. The removal of this oil by pressing has a beneficial effect on the oil still remaining in the cake; the increase in free fatty acids in one sample was only from 3.94 to 5.42 per cent. in 4 months.

D. G. H.

Chemistry of Fat Spoilage. Analytical Differentiation of the Aldehydes Formed. K. Täufel and K. Klentsch. (*Fette u. Seifen*, 1939, 46, 64-66.)—The important rôle of aldehydes in the spoilage of fats by ageing is emphasised. They affect the odour, taste and chemical reactions of the fat, and may be produced by oxidation effects resulting from the influence of heat or ultra-violet light (cf. Schmalfuss, Werner and Gehrke, *Voratspflege u. Lebens. Forsch.*, 1938, 1, 98) or by the decomposition of proteins by proteolytic enzymes. Aldehydes of such origin have been classified into epihydrin aldehydes (which give a positive Kreis reaction) and "free" aldehydes, which are detected by Von Fellenberg's fuchsin-sulphurous acid test (ANALYST, 1925, 50, 245) and determined by means of the quantitative modification of it evolved by Schmalfuss and his co-workers (*loc. cit.*). Since the fuchsin test is a group-reaction (cf. Täufel and Müller, *id.*, 1931, 56, 259), it is necessary in the first instance to be able to distinguish various types of aldehydes, at any rate qualitatively, and in particular, aldehydes of high and low mol. wts. Thus, in Criegee's modification of the test the fuchsin is replaced by Döbner's violet (diamino-fuchsonimonium chloride, a diamino triphenyl methane dyestuff). This is used with sulphur dioxide in the same way as the fuchsin reagent, but the colour produced is dark violet if the aldehydes of low mol. wt. are present (e.g. acetaldehyde) and greenish-blue for the higher aldehydes (e.g. heptyl or nonyl aldehyde). On the other hand, the method is less sensitive than the fuchsin test (cf. Wieland and Scheunig, *Ber.*, 1921, 54, 2527). These authors

also noted that the compound produced by the reaction between the fuchsin-sulphurous acid reagent and an aldehyde differs in its solubility in organic solvents (e.g. amyl alcohol or chloroform) according to the mol. wt. of the aldehyde. Thus, the compounds of the following aldehydes are red and insoluble in chloroform:—formaldehyde, acetaldehyde, propyl aldehyde, acrolein (a violet shade, and forms an emulsion in chloroform), epihydrin aldehyde, butyl aldehyde and valeryl aldehyde (violet shade). Capric aldehyde forms a violet-red compound which is soluble in chloroform to give a violet solution, and the following aldehydes form violet or violet-blue soluble compounds which appear blue-violet in chloroform:—heptyl, nonyl, undecyl, lauryl and palmityl aldehydes. The test is carried out as follows:—The sample (5 g.) is distilled with 20 ml. of saturated salt solution and a little pumice in a 300-ml. conical flask, fitted with a splash-trap and a vertical condenser (length, 20 cm.), which delivers into a 10-ml. graduated cylinder. To 10 ml. of the mixed distillate are added 1 ml. of the fuchsin and sulphurous acid reagent, and the mixture is shaken and allowed to stand for 5 minutes. This serves as a preliminary test. An appropriate smaller portion of the distillate is then used for the final test, and diluted to 10 ml. with water. Two ml. of chloroform and 2 ml. of the fresh reagent are then added (in this order), and the mixture is shaken in a 50-ml. separating funnel. If, on standing, the colour is concentrated mainly in the interface, the funnel should be manipulated so as to bring it into the chloroform layer, but in some instances it is helpful to observe the nature and colour of the emulsion. If the colour in the chloroform layer is very strong, this layer should be removed, and the residual liquid shaken with more chloroform. Since a certain amount of fractionation may occur during distillation, the distillate may contain a larger proportion of the more volatile constituents, and it is therefore desirable to collect the 10 ml. Positive results were obtained with 5 g. of paraffin containing 80% of butyl aldehyde or 100% of heptyl aldehyde. Satisfactory results were also obtained with 6 samples of olive oil containing various higher and lower aldehydes, together or singly.

J. G.

Analysis of Chaulmoogra Oils. III. *Hydnocarpus wightiana* Oil.

H. I. Cole and H. T. Cardoso. (*J. Amer. Chem. Soc.*, 1939, **61**, 2351–2353.)—The characteristics of the oil cold-pressed from selected fresh seeds of *Hydnocarpus wightiana* were:—sp.gr. 25°/25° C., 0.9549; free fatty acids (per cent. oleic acid), 2.7; saponification value, 201; iodine value (Hanus), 98.4; (α)_D + 55.0°; n_D^{25} , 1.4799; unsaponifiable matter, 0.25 per cent. Power and Barrowcliff (*J. Chem. Soc.*, 1905, **87**, 884) reported that the fatty acids of this oil consist mainly of hydnocarpic and chaulmoogric acids and they found evidence of a lower homologue having the formula $C_{14}H_{28}O_2$ but were unable to isolate it. They concluded from the high iodine value (140.7) that acids of the linolic or linolenic series were present. The authors have shown (*J. Amer. Chem. Soc.*, 1938, **60**, 612) that no members of these series are present, but that the high iodine value and high optical activity (+ 50.4) are due to the presence of gorlic acid. By the method of analysis previously described (*loc. cit.*) the percentage composition of the total fatty acids was found to be:—hydnocarpic acid, 48.7; chaulmoogric acid, 27.0; gorlic acid, 12.2; oleic acid, 6.5; palmitic acid, 1.8; lower homologues of chaulmoogric acid (alepric,

aleprylic, aleprestic and aleprolic acids with unidentified acids), 3-4. The fatty acids consisted of 84.2 per cent. of solid acids and 15.7 per cent. of liquid acids. The solid acids were separated from the liquid acids by crystallisation from 80 per cent. ethyl alcohol and the two fractions were converted into their ethyl esters and fractionated. The details of the separation are those described in the first paper of the series (*loc. cit.*), the only change being that the two final crystallisations were made with 80 per cent. acetone to prevent the formation of ethyl esters. Alepric acid ($C_{14}H_{24}O_2$), aleprylic acid ($C_{13}H_{20}O_2$), aleprestic acid ($C_{10}H_{16}O_2$) and aleprolic acid ($C_8H_{14}O_2$) are newly discovered homologues of chaulmoogric acid (Cole and Cardoso, *J. Amer. Chem. Soc.*, 1939, **61**, 2349). The presence of an optically inactive unsaturated acid with one double bond was deduced from the iodine value, but attempts to isolate it were unsuccessful. A. O. J.

Fermentation Process in Tea Manufacture. IV. Tea Tannin and its Fermentation Products. C. J. Harrison and E. A. H. Roberts. (*Biochem. J.*, 1939, **33**, 1408-1420.)—Freshly-plucked leaves were extracted with boiling water, and the infusion was extracted in succession with benzene, ether and ethyl acetate. The ethyl acetate extract on being concentrated and treated with chloroform, gave a yield of tannin 20 to 25 per cent. of the quantity of total tannins found by analysis of the original leaves. This had all the properties of a true tannin, its solution being precipitated, for instance, by quinine and lead acetate solution, but by gelatin solution only in the presence of acid and salt. The solution gave green to blue colorations when treated with ferric chloride solution. On hydrolysis with acid, no glucose was produced, but some gallic acid could usually be detected. This tannin obtained by hot aqueous extraction was not, however, the true tannin of the leaf. Extraction with cold 5 per cent. oxalic acid solution or 1 per cent. hydrochloric acid gave a tannin with a much higher specific rotation. Moreover, addition of salt precipitated only 25 per cent. of this tannin, whereas all the tannin obtained by hot aqueous extraction was precipitated by salt. *l*-Epicatechin has been isolated from the leaf, and gallo catechin has been detected therein. The galloyl ester of epicatechin appears to be present in some types of leaf only. The native tannin is probably made up of epicatechin and gallo catechin and their simple condensation products. Condensation of the catechins, resulting presumably from combination between the carbinol group of the pyran and the phloroglucinol nucleus, occurs when their solutions are heated or acidified or made alkaline. Under the last-named conditions a pronounced uptake of oxygen can be observed and the tannin gradually loses its tanning properties. From a study of the oxygen uptake, it is concluded that this autoxidation in alkaline solution is a property of the catechol or pyrogallol nucleus. The fermentation of tea tannin, gallic acid, pyrogallol and catechol by means of the peroxidase of the tea leaf was studied, and the rate of oxygen uptake was found to decrease in that order. During fermentation the decrease in reducing power (as measured by titration with potassium permanganate solution) was more rapid than would have been expected from the rate of oxygen uptake, and the discrepancy is attributed to condensation of the tannins to products of low reducing power. During manufacture oxidation of the tannins is 80 to 90 per

cent. complete, and substances with all the properties of condensed tannins are isolated from the extract of "made" tea by precipitation with acid or salt; they have a low reducing power and are precipitated by gelatin. The material precipitated by acid apparently represents the more highly condensed tannins which are responsible for the dark colour of the infusion. Since too great a proportion of this fraction is undesirable, it follows that "if quality of leaf and conditions of manufacture are reasonably constant the acid-soluble tannin-content seems largely to determine the value of the made tea." See also ANALYST, 1939, 64, 615, 616.

F. A. R.

Determination of the Alkaloids in Preparations of *Berberis vulgaris* and *Berberis agrifolium*. K. Brunner and H. Neugebauer. (*Pharm. Zentr.*, 1939, 80; *J. Pharm. Belg.*, 1939, 21, 575.)—The method used for the determination depends on the fact that quaternary bases in ammoniacal solution are insoluble in ether, whilst tertiary bases are soluble. The quaternary bases are reduced and extracted and phenolic bases are determined in the solution. Two g. of the root bark of *Berberis*, 30 g. of ether and 2 g. of ammonia are shaken together for 5 minutes and left with occasional shaking for 1 hour, after which 2 g. of calcined sodium sulphate are added, and the mixture is shaken and left for 10 minutes. It is filtered into a separating funnel, and the flask and filter are washed twice with 20 ml. of ether. The tertiary bases dissolve in the ether and are extracted by shaking 3 or 4 times with 10 ml. of water acidified with 0.5 per cent. sulphuric acid. Ammonia is then added, the bases are re-extracted with ether, the extract is dried with sodium sulphate, and evaporated to a few ml., a little water is added, and the rest of the ether is evaporated. The amount present is determined by titration with sodium hydroxide in presence of methyl red (1 ml. 0.1N sodium hydroxide solution is equivalent to 0.03041 g. of oxyacanthine). The residue in the flask and that left in the filter are united, the ether is evaporated, 30 ml. of water, 5 ml. of dilute sulphuric acid, 5 ml. of dilute acetic acid and 2 g. of zinc dust are added, and the whole is heated on a water-bath for 2 hours. The supernatant liquid is filtered into a separating funnel, and the residue is heated for 5 minutes with 10 ml. of water and 19 drops of sulphuric acid and filtered. This operation is repeated three times, and the combined filtrates rendered alkaline with ammonia, cooled and extracted with ether. The alkaloids are titrated (dimethyl yellow as indicator) and calculated as berberine. After titration the solution is rendered alkaline by the addition of 1 ml. of sodium hydroxide, and again extracted with 30 ml. of ether. Under these conditions the phenolic bases remain in the aqueous solution. The quaternary bases are titrated, and the difference between the last two titrations gives the phenolic bases.

D. G. H.

Identification of Plasmoquin by a Colour Reaction. A. E. Tchitchibabine and Ch. Hoffman. (*Bull. Soc. Pharmac.*, 1939, 5; *J. Pharm. Belg.*, 1939, 21, 755-756.)—The reaction of Schulemann, Schonhofer and Wingler is not specific for plasmoquin, since related substances such as plasmocide give the same colorations. The following reaction is regarded as specific and very sensitive. A feebly acid solution of plasmoquin in sulphuric acid, when treated with iodic acid, develops a violet-blue colour in dilutions up to 1 in 2,000,000. One g. of plasmoquin is

dissolved in 20 ml. of 10 per cent. sulphuric acid and made up to 500 ml. with water. From this, weaker solutions are prepared down to 1 in 2,000,000. Ten ml. of each of these solutions are mixed with 5 ml. of a 10 per cent. solution of iodic acid and allowed to stand. The colour is developed in the cold in 3 to 5 minutes according to the concentration. With plasmocide a less permanent, redder, and less violet colour is formed more slowly.

D. G. H.

Contraphthisine. Communication from the Rijks-Instituut voor Pharmaco-Therapeutisch Onderzoek. (*Pharm. Weekblad*, 1939, 76, 1301-1302.)—A substance known as "Contraphthisine" has recently been placed on the market as a cure for tuberculosis. The leaflet accompanying it contains a statement by a Dr. Kalle of the University Clinic, Cologne, verifying its suitability for this purpose; it also states that contraphthisine contains "*Drosera rotundifolia*, *Thymus serpylli*, *Vinum absinthium*, Alphyton, *Mel depuratum*, *Saccharum album*, Aqua dist." In the work now described it is referred to as a pale yellow liquid, having an odour similar to that of absinthe and a strongly acid reaction; sp.gr., 1.017; dry solids, 4.30; ash-content, 0.640 g. per 100 ml. The presence of the following substances was established:—potassium, aluminium, sulphate, calcium (in small quantities), invert sugar, saccharin, and approximately 3.4 per cent. of alcohol. It was not possible to identify *Drosera*, *Thymus* and *Absinthium* with certainty, as they have no characteristic constituents apart from plant acids (principally malic and tannic acids) and a little volatile oil. It seems therefore that "Alphyton" is alum, with saccharin to mask its taste.

J. G.

New Colour Reaction for the Identification of Cocaine. M. Pesez. (*J. Pharm. Chim.*, 1939, 30, 200-205.)—A few particles (up to 1 cg.) of the alkaloid are shaken gently in a test-tube with 2 drops of conc. nitric acid and 13 to 15 drops of pure sulphuric acid (sp.gr. 1.84), and the tube is placed in boiling water for 5 to 10 minutes. After cooling, addition of 1 ml. of water produces a canary-yellow colour. The solution is re-cooled, treated with 10 ml. of acetone and cooled again, and 5 ml. of sodium hydroxide solution (about 15 per cent. strength) are added. The tube is then shaken and inverted several times to bring the two liquids in contact. The acetone phase suddenly becomes turbid, while at the junction of the liquids a violet-blue ring is formed. On shaking, the colour of the acetone solution becomes deep azure blue, which slowly changes through violet (3 to 5 minutes) to Bordeaux red (20 to 30 minutes). With 1 mg. of cocaine the blue colour is distinct but fleeting; with a few tenths of 1 mg. a deep rose colour is finally obtained. The colour is specific for cocaine and two other local anaesthetics (delcaine and alypine); hence this reaction should be useful in toxicology. The eucaines give a violet-blue colour, and other local anaesthetics give colours varying from orange-yellow to reddish-brown in the acetone phase. Characteristic colours are also given by some other alkaloids, aromatic hydrocarbons and barbiturates (with phenyl radicals). Benzene gives an intense violet colour, owing to formation of *m*-dinitrobenzene; the reaction can therefore be used for the detection of the nitric ion. Toluene gives a slight pink colour, the xylenes a yellowish-green, and naphthalene a red colour. Normal methyl-ethyl-phenyl-malonylurea (Prominal-Isonal) also colours the acetone solution yellow, but on addition of

soda the colour changes through greenish-yellow and greyish-blue to azure blue, which is stable for a few minutes and then changes slowly through violet to the final Bordeaux red.

E. B. D.

Determination of Bismuth, Iodine and Quinine in Quinine Iodobismuthate and its Injectable Preparations. G. N. Thomis and G. Ph. Kopanaris. (*J. Pharm. Chim.*, 1939, 30, 193–199.)—The complex salt, quinine iodobismuthate, when prepared by the Codex method, has the formula $(\text{BiI}_3)_2\text{C}_{20}\text{H}_{24}\text{N}_2\text{O}_2 \cdot 2\text{HI}$, but the composition varies with the method of preparation. Methods used for the determination of bismuth, iodine and quinine in the salt itself are unsuitable for its oil suspensions or its special aqueous preparations, which may contain other substances, e.g. chlorides, and the following is proposed for these:—(A) *Aqueous preparations*.—These solutions in ampoules usually contain 0.01 g. of bismuth per ml. Ten ml. of the filtered liquid are pipetted into a 150-ml. separating funnel, 2.5 ml. of 15 per cent. sodium hydroxide solution are added, the quinine is extracted by shaking vigorously with (a) 50 and (b) 30 ml. of chloroform, and the extracts are filtered into a weighed dish through a small, dry, defatted piece of cotton in the delivery tube of the separating funnel (length of tube, approximately 3 cm.). After each operation the sides of the funnel are washed 3 times with 4 ml. of chloroform, without shaking the liquids, and the washings are added to the principal extracts. The solution is evaporated on a water-bath and heating is continued until the initial oily residue becomes white and opaque. (This prolonged heating completely removes ethyl carbamate, if present.) The residue is treated twice with 4 to 5 ml. of ether, which is removed on the water-bath, and dried at 105° C. to constant weight. The gravimetric result may be checked volumetrically by dissolving the residue in a known volume of *N*/10 hydrochloric acid and titrating back, with methyl red as indicator, or, if a micro-burette is available, the method may be shortened by the direct titration of the chloroform extract with alcoholic *N*/10 hydrochloric acid, bromophenol blue being used as indicator (*cf.* Thomis, *J. Pharm. Chim.*, 1936, 24, 162; Thomis and Iatrides, *ibid.*, 1935, 21, 585.) Iodine is then determined in the quinine-free solution, to which are added 50 to 60 ml. of chloroform and then, carefully, 5 ml. of nitric acid (sp.gr. 1.4) which has been cooled to about 4° C. The separating funnel is immediately stoppered and shaken vigorously for a few minutes. After clarification the lower layer is run off into a 500-ml. conical flask (the cotton filter having first been removed from the tube), the funnel is washed twice with 5 ml. of chloroform, and the extraction and washing are repeated first with 30 ml. and then with two 5-ml. portions of this solvent. After addition of 1 to 2 g. of pure sodium bicarbonate and 120 to 130 ml. of 95 per cent. alcohol to the combined chloroform extracts, the iodine is titrated with *N*/10 sodium thiosulphate solution. The nitric acid solution, free from quinine and iodine, is decanted or filtered, if necessary, into a 400-ml. beaker, the chloroform is gently boiled off, and the large excess of acid is nearly neutralised with sodium hydroxide solution and, after dilution of the still acid liquid to 200 ml., the bismuth is precipitated with hydrogen sulphide at 20° C. The precipitate is filtered off, washed well with hydrogen sulphide water, alcohol, ether and, finally, re-distilled carbon disulphide

until all free sulphur is removed. It is then dried for an hour at 100° C. and weighed, counterpoised filter papers being used for filtration and weighing. (B) *Solid quinine iodobismuthate*.—Exactly 0.5 g. of the salt is shaken in a 150-ml. separating funnel with 5 ml. of 30 per cent. sodium hydroxide solution until all red particles have disappeared, the salt which finally adheres near the stopcock being detached by water from a washing-bottle. The solution is shaken well for 2 to 3 minutes with 50 ml. of chloroform, 1 to 1.5 ml. of saturated tartaric acid solution is added, and the mixture is again shaken vigorously for 5 minutes and then allowed to stand for 10 minutes. The rest of the determination is as before. (C) *Oily suspensions*.—Ten ml. of the thoroughly mixed suspension are treated with excess of ether, the liquid is decanted through a weighed Schott No. 4 filter with porous glass plate, and the precipitate is washed first by decantation and then on the filter with ether until the washings no longer give an oily residue on evaporation. The precipitate is dried at 100° C. to constant weight and the quinine iodobismuthate in the suspension is calculated. The rest of the analysis is as for (B). *Remarks*.—Analysis of the salt itself showed the presence of an appreciable amount of sodium chloride as impurity. The method is accurate, very simple and rapid (complete analysis requires 3 to 4 hours). E. B. D.

Chemical Assay of Powdered Ergot. C. Daglish and F. Wokes. (*Quart. J. Pharm.*, 1939, 12, 451–467.)—The extraction of the total alkaloids of ergot by different methods was investigated. The method of the British Pharmacopoeia, 1932, employing a single half-hour shaking with alkaline ether at room temperature, did not extract the alkaloid completely, but the efficiency was increased to about 80 per cent. when mechanical shaking was employed. It was further increased in high potency ergots by decreasing the drug/menstruum ratio. The method of Hampshire and Page (*Quart. J. Pharm.*, 1936, 9, 60; cf. *ANALYST*, 1936, 61, 489), employing a single 5-hour extraction, also failed to give complete extraction, even when the amount of ammonia was increased or when dilute ammonia was used. The average efficiency was 78 per cent., but was higher with ergot of low potency. In both methods the reduced efficiency is due to incomplete extraction, since more alkaloid could be obtained by re-extraction of the marcs. The B.P. method gave slightly higher results than Hampshire and Page's method when both were carried to exhaustion, owing, it is believed, to destruction of some of the alkaloid by heat in the latter method. An improved method of extraction has been devised; it gives an average efficiency of 93 per cent. and yields results not significantly different from those obtained by the B.P. method when both are carried to exhaustion. In this method 5 g. of ergot are shaken mechanically for 30 minutes with a mixture of 49 ml. of acetone and 1 ml. of 10 per cent. ammonia. The acetone extract is filtered, and 40 ml. of the filtrate are mixed with 80 ml. of anaesthetic ether and shaken with four 10 ml.-portions of aqueous 1 per cent. tartaric acid solution. The combined tartaric acid extracts are warmed to remove acetone and ether, cooled, and made up to 50 ml. with tartaric acid solution. One re-extraction of the marc is sufficient to give a reliable figure for the total alkaloids. The authors also show that the experimental errors introduced by the use of a colorimeter are considerably reduced by taking 8 readings instead

of 2, by taking readings against 15-mm. and 20-mm. depths of standard, by comparing solutions differing in colour intensity by not more than 10 per cent., and by eliminating traces of yellow or red colour in the blue colours matched by taking care to remove as much fat as possible from the ergot before beginning the assay.

F. A. R.

Colorimetric Determination of Ergotoxine and Ergometrine. S. A. Schou and J. Bennekou. (*Dansk Tidsskr. Farm.*, 1938, 12; *J. Pharm. Belg.*, 1939, 21, 477-478.)—It is shown that the colour reaction given by these alkaloids with *p*-dimethylaminobenzaldehyde and described in the British Pharmacopoeia, 1932, and the modified reaction described by Allport and Cocking (*Pharm. J.*, 1932, 129, 235; *Abst., ANALYST*, 1932, 57, 725) are identical and may be used for quantitative determinations with the Pulfrich photometer. When an aqueous solution of the alkaloids of *pH* 6 is shaken with ether, ergotoxine passes completely into the ethereal layer, and 90 per cent. of the ergometrine remains in the aqueous layer. If the ethereal layer is shaken with a fresh portion of water of *pH* 6, 9/10ths of the ergometrine that it contains is removed, so that an almost complete separation of the alkaloids is effected.

A. O. J.

Biochemical

Effects of Sulphur Dioxide on Vegetation. G. S. Whitby. (*Chem. and Ind.*, 1939, 58, 991-997.)—The work described originated in complaints of damage, due to sulphur dioxide from the largest non-ferrous smelter (dealing with concentrates of lead and zinc sulphides) in the British Empire, at Trail, British Columbia. The emission of this gas in the peak year (1930) amounted to about 550 tons per day, and, owing to the situation of the works (in a deep valley), the problem became so acute that an International Joint Commission was appointed to investigate it (see also M. Katz *et al.*, "*Effect of Sulphur Dioxide on Vegetation*," Ottawa; National Research Council, 1939). The main conclusion reached by the Commission was that the damage is trifling so long as the following conditions are observed:—If at any time at a point 7 miles south of the smelter the concentration of sulphur dioxide in the air exceeds 1 p.p.m. for 3 consecutive 20-minute periods, the emission of the gas should be reduced until the concentration at the point in question falls to 0.5 p.p.m. Most of the argument in connection with the Trail complaint centred on the lower concentrations, since it was contended that concentrations of the gas that produced no visible markings on vegetation could, nevertheless, cause injury. Climatic factors, which, as a rule, favour the occurrence of the gas in high concentrations for relatively long periods of time, include low winds, low temperatures, high humidities and, especially, fog or mist. The sulphur dioxide concentrations were recorded continuously by aspirating air through a conductivity cell containing very dilute hydrogen peroxide, which converted the gas into sulphuric acid; the corresponding increase in conductivity was obtained by means of an automatic recording Wheatstone bridge (*cf.* Thomas, *Ind. Eng. Chem., Anal. Ed.*, 1932, 4, 253). After 20 minutes the air was automatically diverted to a second similar cell for the next 20 minutes, so that the first cell could be renewed. The investigations showed that the sulphur-content of the

surrounding vegetation can be abnormally high at distances from the smelter greater than those at which visible injury to field crops and trees occurs; this is because the sulphur-content is influenced by both the time of exposure and the concentration of the gas, and because in some instances (*e.g.* the needles of coniferous trees) the increase in the sulphur-content is cumulative. Fumigation experiments indicated in fact that, as a rule, the sulphur-content may rise to 4 times the normal value without producing any injury; this probably is because small doses of the gas are assimilated and oxidised to sulphate, whereas heavy doses extending over a short period may result in damage without raising the sulphur-content appreciably (see below). The pH and exchangeable-base content of the soil near the smelter (especially in the top 1 inch) were lower than the normal values, but this reduction fell off rapidly at greater distances from the smelter, being almost negligible 8 miles away, although injury to the leaves of plants was then apparent. The sulphate-contents of the water-supplies in the area were not affected, but seemed to depend rather on the nature of the formation through which the water had seeped. Studies of the growth-rates of 10,000 trees (Douglas fir and yellow pine) by comparisons of the widths of the annual rings of trees, inside and outside the area, showed that, whilst the growth rates outside the area could usually be correlated closely with the rainfall, those inside did not respond to this form of stimulation. Experiments with 43 groups of conifers (450 trees) showed that they were far more susceptible to injury at the time of active growth than during periods of dormancy. Transplanted conifers (and especially the larch) were more susceptible than conifers in their natural habitat, and seedlings were less resistant than the older trees. The importance of humidity and of the intensity of the sunlight are illustrated by experiments with barley and alfalfa, which showed that, in general, all factors that favour the opening of stomata and a condition of leaf turgor (corresponding with active photo-synthesis) promoted absorption of sulphur dioxide and, consequently, injury. The moisture-content of the soil also influences leaf turgor, and it was found that wilting plants were more resistant in a dry than in a wet soil. Photo-synthetic activity also varies with the age of the plant, and, consequently, barley was found to be most susceptible when young and alfalfa at middle-age. Measurements of the stomatal openings by three different methods enabled the effects of sulphur dioxide to be traced. Thus, continuous fumigation with 0.40 p.p.m. of gas was without effect for the first 4 days, but subsequently the stomata opened to only 65 per cent. of the normal extent; just prior to this, signs of acute injury were first evident, and after 1 week this was apparent in 27 per cent. of the leaves. Barley is the most sensitive of the cereals, although less sensitive than alfalfa; yet it was shown that continuous exposure of the latter to a low concentration of gas (*e.g.* 0.30 p.p.m.) for a long period (*e.g.* 66.5 hours) injured less than 1 per cent. of the leaves and did not affect the yield. As a rule the chemical composition of the plant was unaffected, although when injury was severe there was some reduction in the contents of sucrose and polysaccharides of alfalfa. The assimilation and respiration of carbon dioxide were measured by passing the air entering and leaving the fumigation cabinets through sintered glass plates in a conductivity cell containing 0.0052 *N* sodium hydroxide solution and a little butyl alcohol (to prevent foaming). If the contents of the cell were

renewed every 2 minutes, the changes in conductivity indicated the carbon dioxide concentration with a sensitiveness of 1 p.p.m. Such measurements were found to be a much more sensitive index of the effects of the gas than the appearance of visible markings, especially where short exposures to high concentrations were concerned (see above); thus, a shock-effect is produced, followed by a recovery without necessarily any visible signs of injury. In general, up to 0.5 p.p.m. was without effect on the photo-synthesis of alfalfa, but when this concentration was exceeded reduced assimilation and increased elimination of carbon dioxide in the day and night, respectively, resulted; this effect corresponds with that produced by a sudden reduction in the intensity of the light. Since recognisable injury to the leaves seldom followed such treatment, it is concluded that sulphur dioxide may inhibit temporarily the chlorophyll mechanism without actually injuring the chlorophyll itself or the protoplasm.

J. G.

Method of Ashing Soft Tissues Preliminary to the Determination of Cations. M. V. Buell. (*J. Biol. Chem.*, 1939, 130, 357-363.)—The two methods most frequently used for ashing tissues are ignition in a muffle furnace and wet digestion with sulphuric acid. There are serious objections to both, and a method has now been developed for digesting the dried tissue, freed from fat, with nitric and perchloric acids at a low temperature. A sample of the moist tissue, weighing 10 to 15 g., is minced and introduced into a tared 140-ml. stout conical flask (Pyrex) fitted with a standard ground-glass joint. The flask is reweighed, 15 ml. of pure acetone are added, and the mixture is thoroughly stirred with a rod. The flask, with the rod, is then placed on a steam-bath until the acetone has evaporated, after which it is transferred to a cold electric oven, and the temperature is raised slowly to 105° C., and maintained at that level for 24 hours. The mixture is ground in the flask with the rod, which is then removed, and the flask is re-weighed, the loss representing the moisture-content of the tissue. The lipids are removed by extracting the tissue successively with 20-, 10- and 10-ml. portions of warm anhydrous ether and three 10-ml. portions of warm low-boiling petroleum spirit. The tissue is dried as before and re-weighed. Ten ml. of water and 10 ml. of conc. nitric acid are added to the dry fat-free residue, the flask is placed on an electric hot plate covered with a sheet of asbestos, and the low heat is turned on. After the volume has been reduced to about 10 ml. and the tissue has dissolved (1 to 2 hours), 3.5 ml. of 70 per cent. perchloric acid are added, and the heating is continued until ashing is complete and the residue is almost dry (20 to 24 hours). As the residue approaches dryness, the sides of the flask are washed down with a fine jet of water, and the solution is evaporated to dryness. While the flask is still warm 12.5 ml. of water are added from a burette, and a ground-glass stopper is inserted. The ash dissolves slowly but completely, and the resulting solution is suitable for analysis without further treatment. Calcium should be precipitated at pH 4.2 with the aid of bromocresol green, to avoid contamination with magnesium. Phosphates must be removed prior to the determination of sodium, and phosphates and ammonium prior to the determination of potassium. Both results are achieved by adding to the solution in the digestion flask a small drop of 0.1 per cent. alcoholic phenolphthalein solution and just sufficient solid calcium

hydroxide to make the solution definitely **alkaline**. The solution is filtered after standing for an hour. Ammonium ion is removed from an aliquot portion by evaporating to dryness on a hot-plate, and the residue may be used for the determination of potassium. Almost quantitative recoveries of calcium, magnesium, sodium and potassium were obtained when known amounts of these ions were added to beef muscle.

F. A. R.

Colorimetric Semi-Micro-Determination of Arsenic in Urine. J. V. Harispe. (*J. Pharm. Chim.*, 1939, 30, 58-70.)—The author's method for the determination of arsenic in urine is a modification of that of Denigès, in which the arsenic sol is stabilised by silicic acid. It can be used to study the elimination of arsenic after the administration of arsenic-containing medicaments. From 1 to 50 ml. of urine (containing 50 to 1000 mg. of arsenic) are treated in an evaporating dish with 5 g. of crystalline magnesium nitrate and evaporated to dryness on a water-bath, and the dish is then heated for 3 to 4 minutes in a muffle furnace at a dull red heat. After cooling, the residue is moistened with water and taken up in 10 ml. of conc. hydrochloric acid. The liquid is again evaporated on the water-bath and the dish is heated in the oven for a few minutes to destroy traces of nitrites. After cooling, 10 ml. of Bougalt's hypophosphorous and hydrochloric reagent (*J. Pharm. Chim.*, 1907, 26, 13) are added, and the liquid is stirred and treated, drop by drop, with 5 ml. of a solution of 100 ml. of potassium silicate solution (sp.gr. 1.28; SiO_2 , 20.3 per cent.; K_2O , 9.47 per cent.) in 500 ml. of water. The resulting colourless liquid is treated with 1 drop of *N*/10 iodine solution and made up to 20 ml. It is of advantage at this stage to centrifuge the liquid for a few seconds in a perfectly dry tube; this is particularly to be recommended if the colour is later to be determined by a photometric method. The clarified liquid is heated in a dry degreased Pyrex test-tube (conveniently 16 × 180 mm.) in a water-bath for 30 minutes and then allowed to cool in air.

The Standard Colours.—As the presence of mineral salts slightly alters the final colour there is used instead of water in the preparation of the standard colours a solution of 300 g. of magnesium chloride hexahydrate in 180 ml. of water. A quantity of 0.4165 g. of officinal sodium arsenate in the form of transparent non-efflorescent crystals (\equiv 100 mg. of arsenic) is dissolved in the magnesium chloride solution and the volume is made up to 100 ml. with the chloride solution to give the main standard solution. Pyrex test-tubes identical with the first (the diameters must be the same to within 0.5 mm.) are chosen; into eleven of these there are put 0, 0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4, 4.5 and 5 ml. of a 1:10 dilution of the main solution, and into a further five tubes are put 3, 3.5, 4, 4.5 and 5 ml. of a 1:5 dilution of the main solution. The volume in each tube is made up to 5 ml. with the magnesium chloride solution, and there are added to each tube 10 ml. of Bougalt's reagent, 5 ml. of the diluted potassium silicate solution, and 1 drop of *N*/10 iodine solution. The tubes are then heated for 30 minutes in a boiling water-bath. To avoid evaporation of the water which is exuded from the upper surface of the gel by syneresis it is advisable to cover the upper surface of the liquid with a few ml. of vaseline oil after cooling; this reduction of evaporation is assisted by using long narrow test-tubes. Such standard colours are

usable for about a year if the tubes are not shaken and if the colour of the liquid in the bottom part of the tube is used for comparison. The quantities of arsenic in the tubes are 0, 0.05, 0.10, 0.15, . . . 0.50, 0.60, . . . 0.90, 1 mg. Directions are also given for making the determination by means of a photo-colorimeter.

E. M. P.

Relation of Cuprous Creatinine to Tests for Sugar in Urine. M. Samson. (*J. Amer. Chem. Soc.*, 1939, **61**, 2389–2392.)—Creatinine, owing to its own reducing power and its property of dissolving cuprous oxide, is the chief source of trouble in determining glucose in urine. Benedict (*J. Biol. Chem.*, 1908, **5**, 484) believed the solution known by his name to be more sensitive to glucose, either in pure solution or in urine, than is Fehling's solution. This is not correct, as some aqueous glucose solutions that yield typical Trommer or Fehling reactions give only an almost imperceptible haze with Benedict's solution. For urinary work, however, Benedict's solution is unexcelled, since it gives bulky turbidities with concentrations of glucose to which other reagents are not sensitive, and clear solutions with urine containing no glucose but responding to the Trommer and Fehling tests. When 0.1 per cent. of glucose is added to the filtrate obtained after treating normal urine with blood charcoal, the mixture gives with Benedict's test the slight haze given by an aqueous solution of glucose of the same concentration. If, however, Benedict's reagent is added to the untreated urine containing this amount of glucose, an opaque turbidity appears, and this indicates that the charcoal removes a promoter from the urine. Lloyd's alkaloidal reagent, a type of fuller's-earth used by Folin and Berglund (*J. Biol. Chem.*, 1922, **51**, 209; *Abst.*, *ANALYST*, 1922, **47**, 268), has an action similar to that of the charcoal. Of the substances known to be removed from urine by Folin and Berglund's method, creatinine alone was found to be a promoter of the reaction. When the correct amount of creatinine is added to aqueous solutions of glucose they respond to Benedict's test in the same way as urine containing glucose. Experiments on the reduction of Benedict's solution by glucose and creatinine at temperatures below boiling-point showed that at 60° C. a bulky, slightly yellow precipitate and at 50° C. a voluminous white one that agglutinated easily were formed. Analysis of this precipitate proved it to be a compound of cuprous oxide and creatinine containing one atom of copper to one molecule of creatinine, probably the simple addition compound $C_4H_7ON_3 \cdot CuOH$. Although susceptible to oxidation, cuprous creatinine is otherwise very stable, the bond being resistant to alkali. The complex is very soluble in alkali hydroxide solutions, but in sodium carbonate solutions it is insoluble and very voluminous. The sensitivity of Benedict's test and the delay in precipitation from solutions containing alkali hydroxide are therefore explained. It is well known that, in addition to creatinine, there are other substances normally present in urine or resulting from pathological states or derived from administered medicaments which interfere with the course of alkali-copper reactions. Reduction by creatinine has a greater interfering effect in those tests that use a larger proportion of urine to reagent (the Trommer and most variations of the Fehling tests) than in Benedict's test, in which the urine comprises less than 10 per cent. of the mixture. Even in this restricted proportion of urine,

creatinine has interfering properties as well as the desirable sensitising effect. Creatinine promotes precipitation only in a range of concentration from 0.03 to 0.15 per cent.—the range in normal urine. This concentration of creatinine sensitises the Benedict test for the range of glucose (0.05 to 0.3 per cent.) that gives doubtful reactions with other tests. A higher creatinine-content, fortunately not common in urine containing 0.1 per cent. of glucose, delays the appearance of the opacity beyond the 3 minutes' heating on the water-bath that is recommended (Folin and McEllroy, *J. Biol. Chem.*, 1918, **33**, 513; Abst., ANALYST, 1918, **43**, 299), and the 0.4 per cent. creatinine level reported by Folin (*Amer. J. Physiol.*, 1905, **13**, 66) for a starch and cream diet prevents the precipitation entirely, as it would in any other copper reduction test. Concentrations of creatinine below the optimum range, such as may occur after copious water intake, cause the urine containing glucose to act in the same manner as an aqueous solution, and the faint red haze obtained with Benedict's solution would be considered negligible in accordance with the author's instructions. From these considerations it may be possible to devise a more nearly infallible test sensitive to any significant concentration of glucose in urine. A. O. J.

Chemical Estimation, Stability, and Form of Aneurin in Urine. D. Melnick and H. Field, Jr. (*J. Biol. Chem.*, 1939, **130**, 97–107.)—The method of estimating aneurin previously described (*cf.* ANALYST, 1939, **64**, 367–369) has been found to give low results when applied to urine, since the presence of large amounts of salts prevents the complete adsorption of the vitamin on the zeolite. Accordingly, aneurin must first be extracted quantitatively from the urine (for which purpose benzyl alcohol is most convenient), and then transferred back to aqueous solution; the estimation is then carried out as previously described. The urine is collected in a bottle containing 10 ml. of toluene and enough 10 per cent. sulphuric acid (20 ml. for a 24-hour specimen) to maintain the pH at less than 3. The sample is adjusted to pH 5 and concentrated under reduced pressure in a distillation-flask fitted with a tap-funnel containing benzyl alcohol. Anhydrous sodium sulphate (10 g. to a 12-hour sample) is added to the urine to prevent the residue obtained after concentration from becoming gummy, and a few glass beads are added to prevent bumping. When salts begin to separate out, 80 ml. of benzyl alcohol are added, and the concentration is continued until only a few ml. of water remain. The mixture is centrifuged, and the benzyl alcohol layer is removed. The aqueous phase is shaken with more benzyl alcohol (40 ml.) and the mixture again centrifuged. The combined alcoholic extracts are shaken with an equal volume of acidulated water and 4 volumes of ether, and the solvent layer is again extracted with acidulated water. The combined aqueous phases are extracted with ether, and then freed from ether by distillation under reduced pressure. The aqueous extract, now at pH 3.5 to 4.5, is subjected to permutit adsorption and elution by the standard procedure. The entire potassium chloride eluate (10 ml.) is poured into a 100-ml. centrifuge bottle, the adsorption tube is washed with an equal volume of 95 per cent. ethyl alcohol containing 50 mg. of phenol, and the washing is added to the eluate. A little thymol blue is added to the mixture, followed by sodium hydroxide solution (40 per cent. solution at

first, then *N* solution for the final adjustment) dropwise until a faint blue colour appears. Immediately, 20 ml. of the diazotised *p*-aminoacetophenone reagent (Prebluda and McCollum, *cf.* ANALYST, 1939, 64, 366) are added, and the bottle is stoppered and allowed to stand overnight. The standard solution is prepared in similar fashion with 50γ of aneurin in 0.5 ml. of acidulated water added to 9.5 ml. of a blank potassium chloride eluate. On the following day the coloured vitamin derivative is extracted with xylene, and the colour of the xylene layer from the unknown sample is compared with that from the standard in a micro-colorimeter. The recovery of added aneurin was about 90 per cent., and duplicate experiments gave results that varied by less than ± 5 per cent. The failure to obtain complete recovery is attributed to the presence of salts not completely removed by the benzyl alcohol treatment. Almost theoretical recovery was obtained by using smaller aliquot portions of urine. The method appears to be specific for aneurin, and numerous drugs were found to give no interfering colour reactions. The vitamin in the urine, preserved in the way recommended, is stable for at least a month. The vitamin is excreted as free aneurin, no phosphorylated material having been detected.

F. A. R.

Semi-Micro-Estimation of Amino Acids. H. R. Ing and M. Bergmann. (*J. Biol. Chem.*, 1939, 129, 603–607.)—An apparatus was designed for the estimation on a semi-micro scale of amino acids in protein hydrolysates by precipitation as sparingly soluble salts. An essential requirement of the method is that the mixture of amino-acids and reagent should be agitated and the amino-acid salt filtered under temperature conditions as constant as possible. A correction for the impurities present in the mother liquor that adheres to the precipitate and filter has to be applied, since the salt cannot be washed. The reaction mixture is placed in a small bottle (in one apparatus of 5 ml., in another of 1.5 ml. capacity), upon the neck of which rests an inverted sintered glass micro-funnel of appropriate size. Both are weighed at the beginning of the test. The whole is enclosed in a suitable centrifuge tube (turned upside down), which is closed by a tightly fitting rubber stopper. Several of these tubes are suspended vertically, stopper downwards, in an ice-bath in a suitable apparatus which rotates backwards and forwards. After being thus agitated for 24 to 48 hours, the centrifuge tubes are removed from the bath, dried and immediately inverted into large centrifuge pots packed with crushed ice and water, the tubes being held vertically by cork collars. In each the small bottle containing the reaction mixture now rests upside down on the filter, and by centrifuging for 5 to 10 minutes at 2500 to 3000 r.p.m., filtration is rapidly and completely effected, the filtrate collecting in the bottom of the centrifuge tube. The bottle and filter are weighed immediately and re-weighed after being dried in a desiccator to constant weight. The loss of weight on drying enables a correction to be made for the solids contained in the mother liquor adhering to the precipitate and apparatus, provided that the total solids present in the original reaction mixture is known. Thus from the uncorrected weight (m_2) of the precipitate, a weight equal to $x(m_1 - m_2)/(v - x)$ must be subtracted, where x is the loss of weight in g. on drying and m_1 is the total solids originally present in the volume v ml. of solution used. The correction

can, of course, be checked by washing the dry amino-acid salt on the filter with an ice-cold saturated solution of the pure amino-acid salt. Examples are given of the estimation of glycine and alanine with sodium dioxypyridate, leucine and arginine with naphthalene- β -sulphonic acid, and proline with ammonium rhodanilate. In no instance did the observed value differ by more than 2 per cent. from the theoretical value.

F. A. R.

Distribution of the Fatty Acids in Halibut Intestinal Oil, with a Note on the Presence of Free Fatty Acids in the Intestines of Fish. J. A. Lovern and R. A. Morton. (*Biochem. J.*, 1939, 33, 1734-1739.)—If the suggestion previously advanced is correct (*cf.* ANALYST, 1939, 64, 444), that intestinal vitamin A assists in the process of fat absorption in fish, then one would expect to find vitamin A esterified with all the fatty acids concerned, the amounts of the various esters present being in the same proportion as the individual fatty acids in the fat. To examine this suggestion, a large quantity of halibut intestines was extracted with ether, and the resulting oil was first separated by treatment with acetone into soluble "fat" and insoluble "phosphatides." The "fat" contained 73 per cent. of free fatty acids, 6 per cent. of vitamin A and 21.5 per cent. of unsaponifiable matter. Free fatty acids were next (incompletely) removed by treatment with alkali, then the residual oil was freed from cholesterol by cooling the acetone solution, and finally the liquid fraction was subjected to molecular distillation. The acid components of the various distillation fractions were analysed by standard methods, and the composition so derived was compared with the compositions of the fatty acid component of the undistilled neutralised oil, of the free fatty acids and of the phosphatides obtained in the same way. The neutralised oil had substantially the same fatty acid composition as the fatty acids washed out from it, but the phosphatide fatty acids were appreciably different. The composition of the fatty acids of the high-boiling fraction, presumably containing most of the vitamin A esters, was also very similar to that of the free fatty acids. Thus the evidence supports the hypothesis that vitamin A assists in fat absorption processes; it seems unlikely that the phosphatides are directly concerned in the process. Examination of freshly caught fish showed that appreciable quantities of free acid are present in the living gut.

F. A. R.

Vitamin A Content of Cheese. A. W. Davies and T. Moore. (*Biochem. J.*, 1939, 33, 1645-1647.)—A sample of English Cheddar cheese was examined for vitamin A and carotene by the colorimetric method. Vitamin A, as determined by the antimony trichloride test applied to the unsaponifiable fraction, was present in an amount equivalent to 3 I.U. per g., and carotene, determined by the tintometer, in an amount equivalent to 6 I.U. (3.5 γ) per g. or 3 I.U. (1.8 γ) per g. when the estimation was carried out on the unsaponified fat and on the unsaponifiable matter respectively. Thus the total vitamin A activity of the cheese was 6 to 9 I.U. per g. or 18 to 27 I.U. per g. of fat. This value was confirmed by biological assay, which indicated a potency of about 7.5 I.U. per g.—a value in agreement with the potency of an average milk-fat. Values of the same order were obtained by the colorimetric method with other cheeses made from whole milk, namely,

Camembert, Cheshire, Empire red, Empire white, Gruyère and Stilton, but smaller amounts of vitamin were found in cheeses of lower fat-content such as Dutch Edam and Danish blue.

F. A. R.

Seasonal Variations in the Vitamin A Content of Certain Visceral Organs of the Geelbek or Cape Salmon (*Atractoscion aquideus* C. and V.). C. J. Molteno and W. S. Rapson. (*Biochem. J.*, 1939, 33, 1390–1393.)—The liver oil and the visceral oil (*i.e.* the combined oils from the pyloric caeca and the intestines) of female geelbek were obtained by extracting the minced tissues with ether after desiccation with anhydrous sodium sulphate. Samples were examined each month between November and May of the following year, fish weighing between 12 and 15 lb. being selected. As far as possible, the fish in each month's sample had livers of approximately the same size. The vitamin A content of each oil was measured spectrophotometrically, and the iodine value was also determined. Fish caught in November had small livers with a very low oil content. This oil had a very high vitamin A content, equivalent to nearly 25 per cent. of the weight of the oil, and a low iodine value. As the season advanced and the fish began to feed intensively the liver increased in size and in oil-content, but the concentration and the total amount of vitamin A in the liver oil decreased, and the iodine value increased. Thus the fall in the concentration of vitamin A cannot be accounted for simply by infiltration of fat. About February or March the amount and concentration of vitamin A in the liver oil began to increase, the iodine value fell, and the liver decreased in size. The visceral oil, on the other hand, contained only a small amount of vitamin A in November, and until May this gradually increased to a high value. The diet of the geelbek during the period of intensive feeding is a very fatty one, and the observation that the vitamin A content of the viscera increases during this period is believed to support the suggestion made by Edisbury, Morton, Simpkins and Lovern (*cf. ANALYST*, 1938, 63, 358) that vitamin A assists in the process of fat assimilation in certain fish.

F. A. R.

Effect of Soil Treatment on the Vitamin B₁ Content of Wheat and Barley. P. C. Leong. (*Biochem. J.*, 1939, 33, 1397–1399.)—Five specimens of flour, obtained from wheat grown on plots receiving respectively no manure, dung, a complete mineral manure, ammonium sulphate only and a complete mineral manure mixed with ammonium sulphate, showed no significant differences in vitamin B₁ content. The values obtained by the bradycardia method were 1.0 to 1.3 I.U. per g. Flour from barley grown on plots similarly treated also showed no significant differences, except that in the 1935 crop a higher vitamin B₁ content (2.0 I.U. per g.) was found in the flour from the plot treated with dung. The other specimens gave values of 0.8 to 1.3 I.U. per g.

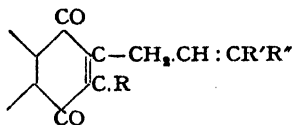
F. A. R.

Factors Preventing Oxidation of Ascorbic Acid in Blood Serum. E. M. Mystkowski and D. Lasocka. (*Biochem. J.*, 1939, 33, 1460–1464.)—The atmospheric oxidation of ascorbic acid to dehydroascorbic acid is catalysed by copper, and the reaction is inhibited by substances that form complexes with copper. Serum globulin has now been found to have such an effect, but not to such an

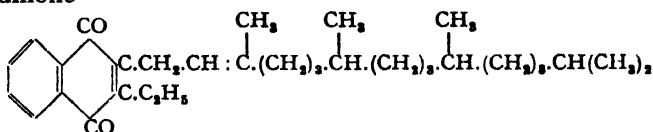
extent as might be anticipated. This is ascribed to the presence, in the globulin, of protein-copper complexes that retain a certain catalytic activity. Casein had a higher inhibitory action than serum globulin, and gelatin a somewhat lower activity than casein. The chloride ion appears to be of particular importance in inhibiting the oxidation of ascorbic acid, and its action is independent of the nature of the cation, and so may be exerted in the tissues and in the serum. The fluoride ion is inactive. (Cf. Høygaard and Rasmussen, *Nature*, 1938, **142**, 293.)

F. A. R.

Blue Alkali Salts of α -Phylloquinone (Vitamin K_1) and Similar Compounds. P. Karrer. (*Helv. Chim. Acta*, 1939, **22**, 1146-1149.)—Treatment of solutions of α -phylloquinone (vitamin K_1) or of vitamin K_2 with sodium alcoholate solutions leads to the formation of an intense blue colour, similar to that found by C. Liebermann (*Ber.*, 1898, **31**, 2903; 1899, **32**, 260, 916; F. Michel, *Ber.*, 1900, **33**, 2402; Liebermann and Lanser, *Ber.*, 1901, **34**, 1543). Analogy with these Liebermann compounds, the structural formulae of which are quoted in this paper, lead to the suggestion that vitamin K contains the group



in its molecule; this was confirmed by MacCorquodale, Binkley, Thayer and Doisy (*J. Amer. Chem. Soc.*, 1939, **61**, 1928), who suggested as the probable formula for α -phylloquinone—



E. M. P.

Isolation of α -Spinasterol from Alfalfa. E. Fernholz and M. L. Moore. (*J. Amer. Chem. Soc.*, 1939, **61**, 2467-2468.)—During experiments to isolate vitamin K from alfalfa, small amounts of a crystalline sterol of m.p. 168° C. were obtained. It was identified as a sterol by the formation of a digitonide, and by the Liebermann, Salkowski and Tortelli-Jaffé reactions, and its properties suggested that it might be identical with α -spinasterol first isolated from spinach (Hart and Heyl, *J. Biol. Chem.*, 1932, **95**, 311), and subsequently found in senega root (Simpson, *J. Chem. Soc.*, 1937, 730). There is little doubt that medicagosterol II, isolated by Dam *et al.* (*Helv. Chim. Acta*, 1939, **22**, 313) is α -spinasterol. The substance found in alfalfa yielded an acetate with m.p. 183° C. and a dinitrobenzoate with m.p. 195° C. and, when the difficulty of purifying it is taken into account, its properties were found to agree closely with those found by Dam (*loc. cit.*) and by Larsen and Heyl (*J. Amer. Chem. Soc.*, 1934, **56**, 2663). A study of its catalytic hydrogenation provided further evidence that it is a sterol, one molecule of hydrogen being absorbed in the presence of Adams's catalyst. The resulting compound was still unsaturated, and the m.p. and $(\alpha)_D$ of the dihydrosterol and its acetate agreed well with those reported for the corresponding compounds prepared from α -spinasterol. Titration with perbenzoic acid indicated that

3 atoms of oxygen were consumed by the sterol and 2 by the hydrogenated product. Larsen and Heyl (*loc. cit.*) state that the corresponding amounts of oxygen absorbed by their specimen were 2 atoms and 1 atom respectively. It is not rare to find sterols reacting with more perbenzoic acid than the number of double bonds would indicate, and the results quoted do not necessarily suggest the presence of three double bonds. Alfalfa meal, which is probably the most accessible source of the sterol, yielded 0.8 g. of α -spinasterol from 3.8 kg. (this figure is given as 2.8 kg. of dehydrated alfalfa later in the paper). The meal was percolated with hexane, and from the extract (140 g.) the dark brown, waxy, unsaponifiable fraction (28 g.) was separated and purified by solution in hot acetone. The purified product (20 g.) was dissolved in hexane and washed five times in succession with an equal volume of 95 per cent. methyl alcohol saturated with hexane. This residue (11 g.) was dissolved in alcohol, and the crystalline deposit of sterol forming in two days was recrystallised from a mixture of alcohol and benzene. The final product (0.8 g.) melted at 165° C. and after nine more recrystallisations the m.p. was raised to 168° C. When mixed with a specimen of α -spinasterol (m.p. 169° to 171° C.) the m.p. was 168° to 169° C. Ultimate analysis of the compound agreed with the formula $C_{20}H_{46}O_{1.1}H_2O$. The α -spinasterol thus obtained was converted into α -spinasteryl-*m*-dinitrobenzoate, α -spinasteryl acetate, and by hydrogenation into α -spinastenyl acetate, which on saponification yielded α -spinastenol. The ultimate analyses of these compounds agreed with their empirical formulae.

A. O. J.

Vitamin P. H. Scarborough. (*Biochem. J.*, 1939, 33, 1400-1407.)—Vitamin P was defined by Szent-Györgyi as a factor that "brought back the fragile and permeable capillaries to their normal state." The evidence upon which this claim rested was derived from observations made on patients with vascular purpura, thrombocytopenic purpura, infectious diseases, myxoedema and diabetes mellitus, and is regarded by the author as unsatisfactory for deciding whether or not there is a vitamin P. Three substances containing flavanones have now been tested:—crude hesperidin obtained during the desiccation of orange juice, purified hesperidin (m.p. 255° to 256° C.), prepared from it by extraction with pyridine, and "citrin," which appears to be a mixture of eriodictyol glycoside and hesperidin. These substances were given orally, by intramuscular injection or through the rectum, to subjects suffering from multiple vitamin deficiencies. In one individual this was artificially induced by giving a diet containing neither fruit nor vegetables, all food being twice cooked. All the patients showed increased capillary fragility before the flavanone preparations were administered, whether tested by the "positive pressure method," in which fragility is determined in terms of the number of burst capillaries occurring in response to an increased intra-capillary pressure, or by the "negative pressure method," in which fragility is expressed as the amount of suction required to burst a single capillary loop. The administration of ascorbic acid had no effect on the fragility, but all three flavanone preparations increased the capillary resistance to normal. It is claimed that the existence of a factor decreasing capillary fragility is for the first time established with certainty.

F. A. R.

Bacteriological

Preservatives for Preparations Containing Gelatin. L. Gershenfeld and D. Perlstein. (*Amer. J. Pharm.*, 1939, 3, 277-287.)—Two types of gelatin were used in 1 per cent. solutions: (a) Pharmagel A, a pork-skin gelatin from an acid-treated precursor, with 75 mg. of tartaric acid added per g. of gelatin to give pH 3 to 4, and (b) Pharmagel B, a bone gelatin from an alkali-treated precursor, with 0.5 g. of sodium bicarbonate added to give pH 7 to 8. The gelatin solutions, with definite concentrations of each preservative, were inoculated respectively with suspensions of *Staphylococcus aureus*, *B. subtilis*, *Proteus vulgaris*, a pink yeast, *Penicillium glaucum* and a suspension of dust from the pharmaceutical laboratory, free access of air being allowed to each. Controls were run and macroscopic readings and sub-cultures for growth were made at monthly intervals over the 4-month test period. The following preservatives were effective for the acid type gelatin:—sodium benzoate in 0.1 per cent. concentration; thymol (0.1); chlorobutanol (0.5); sodium salicylate (0.1); cresol (0.4); parachlorometaxylenol (0.1); oxyquinoline sulphate (0.1); alcohol (8.0); ethyl hydroxybenzoate (0.15); propyl hydroxybenzoate (0.15); butyl hydroxy benzoate (0.15). For the basic type gelatin the following were effective:—thymol in 0.1 per cent. concentration; chlorothymol (0.1); chlorobutanol (0.5); β -naphthol (0.2); phenol (0.5); cresol (0.4); parachlorometaxylenol (0.1); parachlorometacresol (0.25); alcohol (8.0); ethyl hydroxybenzoate (0.15).
D. G. H.

Estimation of Foreign Organisms in Yeast. K. E. Jensen. (*Internat. Congress Agric. Industries*, July, 1939; *J. Inst. Brewing*, 1939, 45, 500).—Methods are described for the quantitative estimation of small amounts of foreign organisms in pressed yeast. *Oidium lactis*.—The main solution contains 1 per cent. of sodium acetate, 1 per cent. of ammonium chloride and 1 per cent. of agar. To each litre of this are added 10 ml. of 10 per cent. potassium dihydrogen phosphate solution, 10 ml. of 1 per cent. calcium chloride solution, 30 ml. of 1 per cent. magnesium sulphate solution, 1 ml. of 1 per cent. sodium chloride solution and 1 ml. of 1 per cent. ferric chloride solution. Five g. of yeast are diluted to 100 ml. with sterile water, and tests are made with 1 ml., 0.1 ml. and 0.001 ml. of the suspension on the medium in Petri dishes. These are incubated for 4 days at 30° C. and the colonies are then observed. It is claimed that one oidium cell in 500 million yeast cells can be detected. *Mycoderma*.—The main solution described above is used, but the agar is omitted, and 0.5 per cent. of glucose and 0.3 per cent. of "Difco" yeast extract are added. Five ml. of this liquid medium are put into test-tubes, 2.5 cm. in diameter, the measured quantities of the yeast suspension are added, and the tubes are incubated at 37° C. and observed once or twice a day. Those in which no surface film has formed are shaken to promote its formation.

Diagnosis of *Streptococcus mastitis* by Cultural Methods. S. J. Edwards. (*J. Comp. Pathology and Therapeutics*, 1938, 51, 250-263.)—Methods have been devised to facilitate diagnosis of chronic *Streptococcus mastitis* in quarter

and composite milk samples. The results of an investigation of the relative streptococci in whole milk, sediment, and gravity cream from the same milk show that on the average cream contains 3.2 times as many streptococci as the milk, and the sediment from 10 ml. 16.2 times as many as 1 ml. of the milk. The number of streptococci in milk is generally very large in mastitis cases, ranging in the tests recorded from 8000 to 375,000 per ml., and it is suggested that 0.01 ml. of the cream is a satisfactory test inoculum; this is conveniently delivered by means of a standard loop wire constructed so as to present two loops 2.3 mm. in diameter. Direct incubation of milk samples does not compare favourably with cultural examination in blood agar for the detection of *Str. agalactiae*, and does not give reliable results, since non-specific growths are frequently encountered. Crystal violet—aesculin—blood agar is recommended, the loopful of milk being first mixed with a small quantity of sterile normal saline and then well mixed with 10 ml. of melted blood agar before being poured into the plate. For the selective cultivation of *Str. agalactiae* a glucose broth medium containing crystal violet (1 in 1,000,000) and sodium azide (1 in 10,000) and adjusted to pH 6.8 was successfully employed; this medium inhibits the growth of *B. coli*, and the presence of *Str. agalactiae* is indicated by the formation (after incubation) of a large flocculent deposit with complete clearing of the supernatant liquid. Its presence may be confirmed by sub-cultivation on blood agar. The addition of sodium azide to milk in a concentration 1 in 20,000 was found to prevent the multiplication of *B. coli* and other organisms during storage and not to affect the number of *Str. agalactiae*. Instead of samples being left in the ice-chest overnight to obtain gravity cream, by the use of azide they can be kept at room temperature.

D. R. W.

Toxicological

Toxicology of Manganese. A. C. Lemos. (*J. Pharm. Chim.*, 1939, 30, 206-212.)—Experiments on rabbits have shown that soluble manganese salts, when injected or absorbed in large doses, are highly toxic, but manganese dioxide, whether ingested or inhaled, has no immediate toxic effect; the animals treated with it, when killed or dying accidentally after various periods up to 5 months, frequently showed injuries to liver and kidneys. The study of the distribution of the metal in the organism has shown that the impregnation of the principal organs, particularly the brain, increases with the time, but that the amounts in the blood and muscle remain practically constant. Thus, the manganese-contents of various organs, after 2 and 5 months respectively, were (in mg. per 100 g.): heart, 0.27 and 2.5; kidneys, 1.00 and 1.90; lungs, 0.07 and 1.7; brain, 1.07 and 5.8; skin and coat, 1.49 and 3.9. Manganese-contents of the muscles were 0.54, 0.47 and 0.45 for 2, 3 and 5 months, and of blood, 0.25 and 0.26 for 3 and 5 months. For a control animal, the respective results for heart, liver, kidneys, lungs, and skin and coat were not greater than 0.03; brain, 0.67; muscles, 0.01; blood, traces. The manganese-content of small organs (surrenals, marrow, testicles and spleen) was frequently high; this, with the localisation in the nerve centres, may explain the accidents notified among workers exposed to the prolonged absorption of considerable quantities of manganese products.

E. B. D.

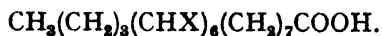
Organic

Wet Combustion Method for Determination of Carbon and of Oxygen Consumed. B. E. Christiansen and J. F. Facer. (*J. Amer. Chem. Soc.*, 1939, **61**, 3001-3005.)—A modified Pettenkofer method was employed (Lunge and Ambler, "*Technical Gas Analysis*," 1934, p. 221). The organic compound (10 to 20 mg.) together with potassium iodate is heated at 190° C. in a reaction vessel with 3 ml. of conc. sulphuric acid for 20 to 40 minutes. The evolved carbon dioxide is absorbed in standard baryta solution, the unused excess of which is neutralised by titration with acid with the use of thymol blue indicator. Oxygen consumed is obtained from a determination of the potassium iodate remaining unused in the reaction vessel, by the usual method of titration with thiosulphate in presence of potassium iodide. S. G. C.

Chromatographic Separation of Palmitic and Stearic Acids from a Mixture of Oleic, Palmitic, and Stearic Acids. C. Manunta. (*Helv. Chim. Acta*, 1939, **22**, 1156-1160.)—When a mixture of saturated and unsaturated fatty acids is adsorbed on magnesium sulphate or, better, on Franconite, the unsaturated acids are found at the top of the adsorption tube, followed by the saturated acids in order of increasing chain length. The following experimental procedure was adopted. A petroleum spirit solution of oleic, palmitic and stearic acids (1 g. of each) was adsorbed in three tubes (3 cm. wide and 60 cm. long) containing magnesium sulphate, $\text{MgSO}_4 \cdot \frac{1}{2} \text{H}_2\text{O}$, previously washed with 100 ml. of petroleum spirit for each tube. Each tube was then washed slowly with 300 to 400 ml. of petroleum spirit, and its contents were divided into six 10-cm. zones, and the corresponding zones of the three tubes were united and washed out with ether. The ethereal solutions were evaporated, and the residues were dissolved in absolute alcohol. After evaporation to 2, 3, or 4 ml. (according to the quantity of material) the alcoholic solutions were chilled and allowed to crystallise, the crystals being filtered off in the cold, washed with a little absolute alcohol and kept in a desiccator. Six fractions consisting of mixtures of palmitic and stearic acids were obtained. Fractions showing similar melting points were united, dissolved in petroleum spirit and again adsorbed on two magnesium sulphate columns. The resulting fractions were again sorted into two lots and once more separated on two columns, 1.5 cm. wide and 36 cm. long. They were thus each separated into three zones. The upper zone of the first consisted of almost pure palmitic acid and the lowest zone of the second lot of fairly pure stearic acid. Similar results were obtained by adsorption on Franconite, the solvent consisting of a mixture of petroleum spirit with 20 per cent. of ether. E. M. P.

Reaction of Wijs' Solution with Tung Oil. S. W. Wan and D. B. Hu. (*J. Amer. Chem. Soc.*, 1939, **61**, 2277-2283.)—In estimating the degree of unsaturation of tung oil with Wijs' solution the presence of elaeostearic glyceride causes much uncertainty because the iodine value thus obtained varies with the experimental conditions (Ho *et al.*, *Ind. Eng. Chem., Anal. Ed.*, 1936, **7**, 96). Various methods for the determination of the percentage of elaeostearic acid in tung oil have been tried but have not given concordant results (Toms, *ANALYST*, 1928,

53, 69; Croxford, *ibid.*, 1929, 54, 445; Bolton and Williams, *ibid.*, 1930, 55, 360; Kaufmann and Baltes, *Fette u. Seif.*, 1936, 43, 93). The properties of elaeostearic acid causing analytical difficulties have been explained by Böeseken *et al.* (*Rec. Trav. Chim.*, 1927, 46, 158, 619; Abst., ANALYST, 1928, 53, 54) by assigning to it the constitution $\text{CH}_3(\text{CH}_2)_3\text{CH}=\text{CHCH}=\text{CHCH}=\text{CH}(\text{CH}_2)_7\text{COOH}$ which according to Thiele's theory of partial valency will absorb halogen (X) in two stages represented by $\text{CH}_3(\text{CH}_2)_3(\text{CHX})_2\text{CH}=\text{CH}(\text{CHX})_2(\text{CH}_2)_7\text{COOH}$ and



With Wijs' solution, X_2 being a molecule of iodine monochloride, the first stage occurs much more rapidly and produces a chloro-compound (Böeseken *et al.*, *loc. cit.*). On the other hand, compounds with non-conjugate double bonds take up the theoretical amount of iodine chloride in a short time, forming iodochloro-compounds. In order to apply the above-mentioned property of elaeostearic acid to the determination of its glyceride in tung oil the kinetics of the slower second stage reaction were investigated. The α -elaestearic acid used was prepared by the method of Thomas and Thomson (*J. Amer. Chem. Soc.*, 1934, 56, 898) and the Wijs' solution was modified to contain exactly equivalent amounts of iodine and chlorine. The iodine values were determined in the usual way with certain refinements in the standardisation and measurement of reagents and thermostatic control of the temperature conditions. The experiments were made with (a) pure α -elaestearic acid and a reaction time of 5 minutes, (b) pure α -elaestearic acid with a reaction time of 2 minutes, (c) pure elaeostearic acid with a reaction time of 10 minutes, (d) pure oleic acid with a reaction time of 10 minutes, (e) a known mixture of α -elaestearic acid and oleic acid with a reaction time of 5 minutes, (f) tung oil with a reaction period of 5 minutes. Similarly experiments were made in which definite volumes of the reaction mixture were withdrawn at definite intervals and the course of the halogen absorption was studied. The reactions investigated were (g) the reaction between α -elaestearic acid and the Wijs' solution in conventional proportions, (h) the same reaction with an equivalent amount of the Wijs' solution sufficient for the first-stage reaction, (i) the same reaction with an excess of the Wijs' solution, and (j) comparison of the halogen absorbed by α -elaestearic acid and by tung oil. With the aid of the results of these experiments a study of the kinetics of the two-stage reaction was made. The values of the velocity constants calculated from the experimental results do not support the assumption that Böeseken's second-stage reaction is either bimolecular or trimolecular. The mechanism suggested by Böeseken is therefore incorrect, and the reaction is most probably a combination of two simultaneous bimolecular reactions—one between the acid and the iodine liberated in the first stage, and the other between the acid and iodine monochloride. The compounds thus formed are $\text{CH}_3(\text{CH}_2)_3\text{CHClCHClCHClCHClCHCl}(\text{CH}_2)_7\text{COOH}$ and $\text{CH}_3(\text{CH}_2)_3\text{CHClCHClCHClCHClCHClCHCl}(\text{CH}_2)_7\text{COOH}$ respectively, the product of the first stage being a tetra-chloro compound. The difference in halogen absorption between α -elaestearic acid and oleic acid has been applied to the analysis of a mixture of these acids and to the determination of elaeostearic glyceride in tung oil, and further work is being undertaken for improving the

latter method. The increase of the iodine value of the oil with increase of temperature or time of contact with the reagent is explained. The absorption of chlorine from iodine monochloride in the first stage explains the requirement of a minimum excess of Wijs' solution. The slight decrease of iodine value with increased amount of oil when the same amount of reagent is used may be accounted for by the fact that a relatively larger amount of α -elaeostearic glyceride absorbs a correspondingly larger amount of iodine monochloride in the first stage, and leaves a smaller excess of halogen which decreases the rate of absorption in the second stage.

A. O. J.

Determination of the Eugenol-content of Essential Oils. P. A. Rowaan and J. A. Insinger. (*Chem. Weekblad*, 1939, 36, 642-643.)—The effects of extraction with solutions of potassium and sodium hydroxide of various strengths and at temperatures ranging from 20° C. to 100° C. have been compared, and the following procedure is recommended:—Ten ml. of the dried and/or filtered oil are pipetted into a 100-ml. flask, the neck of which is graduated from 0 to 10 ml. in 0.1-ml. divisions. To this are added 80 ml. of 1.0 *N* potassium hydroxide solution (A.R. quality), and the flask is then shaken well at intervals of 5 minutes for 30 minutes. Sufficient of the potassium hydroxide solution is then added to bring the level of the unabsorbed oil up into the neck of the flask, where its volume may be read to within 0.05 ml.; the reduction in volume corresponds with the eugenol originally present. Satisfactory results were obtained with oils of clove, pimento, cinnamon leaf and New Guinea lawang oil (an oil from the bast of a plant of the *Cinnamomum* species); bay oil, however, gave unreliable results, owing to the presence of other phenolic substances (e.g. chavicol). If an emulsion is formed during the shaking operation, the oil should first be shaken with a little tartaric acid solution, which is subsequently removed, and the residual oil is dried with anhydrous sodium sulphate and then filtered.

J. G.

Boric Acid Colour Reaction of Flavone Derivatives. C. W. Willson. (*J. Amer. Chem. Soc.*, 1939, 61, 2303-2306.)—Lemon juice evaporated with boric acid produces a brilliant yellow colour and the substance causing the colour reaction has been identified as a constituent of Szent-Györgyi's citrin (Armentano *et al.*, *Deutsch. Med. Wochschr.*, 1936, 62, 1326). The reaction will detect 0.004 mg. of citrin or 0.002 mg. of quercitrin in 0.5 ml. of solution. An attempt to determine the specificity of the reaction was made. Several representative flavones and flavanones were purified by recrystallisation from alcohol, and hydroxychalcones were obtained by the action of alkali on naringin and hesperidin. The reagent used for the determination of the reactivity of these compounds with boric acid was made by mixing equal parts of a saturated solution of boric acid in absolute acetone and a 10 per cent. solution of anhydrous citric acid in absolute acetone. The separate solutions are stable, but the mixture is stable only for one day. To determine the reactivity, the flavone derivative (0.5 mg.) was dissolved in about 1 ml. of dry acetone and the solution was divided into two parts. To one part about 2 ml. of the boric acid and citric acid mixture were added, and the other part was diluted to the same volume with a mixture of equal parts of acetone and the citric acid and acetone solution. After a few minutes the colours of the two

tubes were compared, a stronger colour in the tube containing boric acid being regarded as a positive reaction. By comparing the reactions of the aglycones with the reactions of two glycosides of quercetin, *viz.* isoquercitrin and quercimeritrin it was established that neither the presence or absence nor the position of the sugar group influenced the colour reaction. Of all the flavone derivatives tested, the flavanones gave no colour reaction with boric acid, whereas all the flavones except fisetin gave a reaction. When the lack of reaction of fisetin is compared with the reactivity of quercetin, the importance of the hydroxyl (or other auxochromic) group in the 5-position is evident. The reactivity of kaempferol compared with the failure of naringenin leads to speculation whether the hydroxyl group in the 3-position or the double bond between the 2- and 3-carbon atoms is responsible for the difference, and the opening of the pyran ring to form the reactive chalcone establishes the importance of the double bond and indicates that the pyran ring is unnecessary for colour formation. The configuration of the chalcones suggests that of curcumin, which reacts with boric acid to give a pink colour. If this colour formation is comparable with the flavone reaction, the benzene ring near the ketone (or quinone) group is unnecessary, provided that an auxochromic group is attached to the second carbon atom from the C = O group. The

indicated configuration for reactivity is

$$\begin{array}{ccccccc}
 & a & & & O & & \\
 & | & & & || & & \\
 R - & C & - & C & - & C & = C - R' \\
 (v) & (w) & (x) & (y) & (z) & &
 \end{array}$$

which *a* is an auxochromic group which may probably be = O, OH, OCH₃, etc., and in which R, C(v) and C(w) may form a benzene ring, and C(x), C(y) and C(z) may form part of a pyran ring. In all the compounds examined, R' has been a benzene ring containing either methoxyl or hydroxyl groups. When the pyran ring of fisetin is opened by treatment with alkali, the formation of the hydroxyl group in the ortho position on one of the benzene rings fulfils the requirement of the position of the auxochromic group, but the double bond between carbon atoms on the other side of the ketonic group is missing. A compound of this type may exist in an enolic form (Buswell, Rodebush and Roy, *J. Amer. Chem. Soc.*, 1937, 59, 1767) having a double bond in the required position. Another possible explanation is that in the alkali treatment of fisetin other secondary compounds formed are responsible for the colour reaction. The following substances which may occur in nature with the flavones did not give the colour reaction:—tannic acid, coumarin, coumarinic acid, dextrose and sucrose and the products of their treatment with alkali, phloroglucinol, quinhedrone and salicylic acid. A. O. J.

Pectic Substances of Plants. VI. Relation between Jelly Strength, Viscosity and Composition of Various Pectins. E. W. Bennison and F. W. Norris. (*Biochem. J.*, 1939, 33, 1443-1451.)—The jelly strengths of various pectins were measured by a modified form of the pectinometer described by Buston and Nanji (*Biochem. J.*, 1932, 26, 2090) and viscosities by an Ostwald viscometer. An attempt was made to correlate these two properties with the chemical composition of the preparations. Little relation could be found, although, in general, a high urone content is associated with a satisfactory jelly strength.

Methoxyl-content is not a criterion of jelly strength, as has sometimes been maintained, for first, esters of pectolic and pectic acids prepared by methylation failed to give jellies, and secondly, the methoxyl-contents of prepared pectins bore no relation to their power of forming jellies. The method of preparation is an important factor, and autoclave extraction in presence or absence of sucrose inhibits jelly formation. The jelly strength and viscosity of the same pectin are closely related, and both are reduced when the pectin is heated. Although the ability of pectins to form jellies depends to some extent on their composition, it is primarily determined by molecular size, as indicated by viscosity. Any treatment tending to disaggregate the polygalacturonide chain tends to cause loss of jelly strength (*cf.* ANALYST, 1939, 64, 000). F. A. R.

Inorganic

Determination of Aluminium Oxide in Metallic Aluminium by Means of Cupric Ammonium Chloride. T. Nakamura and S. Yamazaki. (*J. Soc. Chem. Ind., Japan*, 1939, 42, 296-297B.)—A 10-g. sample of aluminium is allowed to react with a solution of 240 g. of cupric ammonium chloride, added little by little to prevent violent interaction. The liquid is finally heated, and the residue of aluminium oxide is filtered off and washed with 1 per cent. hydrochloric acid to remove basic copper compounds as far as possible. The remainder is ignited and fused with potassium bisulphate, and the aluminium in the melt is determined by the usual procedure, the weight being calculated as aluminium oxide. The amounts of aluminium oxide found by this method in virgin ingots ranged from 0.01 to 0.06 per cent. The authors satisfied themselves (1) that anhydrous alumina is practically unattacked in the process; (2) that the aluminium in association with elements such as silicon is almost completely dissolved. S. G. C.

Reaction of Gallium with Perchloric Acid. L. S. Foster (*J. Amer. Chem. Soc.*, 1939, 61, 3122-3124.)—Metallic gallium, which dissolves very slowly in the common mineral acids, has been found to dissolve vigorously in hot perchloric acid (72 per cent. strength); 5 g. of gallium (form not stated) dissolved in 60 ml. of the perchloric acid in 1 hour. The gallium perchlorate produced is soluble in the hot acid liquid, but separates almost completely on cooling, in the form of coarse white crystals of the hexa-aquo salt. A mixture of two-thirds conc. sulphuric acid and one-third perchloric acid was more rapid in solvent action than perchloric acid alone. S. G. C.

Detection and Determination of Germanium with Hydrogen Selenide. V. J. Kouznecov. (*J. Ob. Chem.*, 1939, 9, 1049-1054.)—Hydrogen selenide reacts with quadrivalent germanium in mineral acid solution to form a yellow-orange precipitate; this reaction is specific. When the concentration of germanium is as low as 1 in 10^{-5} a yellow turbidity is produced almost instantaneously, but as a red precipitate of selenium is formed after a few minutes, the test is doubtful for such low concentrations. Owing to this ease of oxidation and to the poisonous nature of hydrogen selenide, an organic derivative which it forms with formaldehyde is used instead of the selenide itself. This compound is produced in a colourless

solution when hydrogen selenide is passed into a solution of formaldehyde which is just acid. It is fairly stable in air, and concentrated solutions become turbid only after standing many hours (sometimes days) in an open test-tube, after which a yellow precipitate is slowly deposited on the sides (the red precipitate of selenium is not formed). In dilute solution the formaldehyde compound causes turbidity after an hour. Its reactions with metallic ions are the same as those of hydrogen selenide solution. With germanium a curdy yellow precipitate, somewhat less reddish and more stable to light than germanium selenide, is formed. It is insoluble in conc. acids (even in hot sulphuric acid) but readily soluble in alkalis. The reaction is sensitive for 0.2 p.p.m. of germanium, and silicic, hydrofluosilicic, hydrocyanic and other acids do not interfere with it. The test is made by adding 2 or 3 drops of the solution prepared as described below to 5 ml. of the solution examined, which contains approximately 1 ml. of conc. hydrochloric acid (the degree of acidity is important). *Preparation of reagent.*—Twenty g. of selenium and 14 g. of aluminium powder are ignited in a closed crucible, with addition of 20 g. of ignited sodium chloride to moderate the reaction. From 5 g. of the impure aluminium selenide thus formed, hydrogen selenide is liberated, in a small flask, by the action of water from a dropping funnel; finally, addition of dilute hydrochloric acid liberates hydrogen (from the excess of aluminium) which sweeps out the remaining hydrogen selenide. The gas is passed at room temperature, with thorough shaking, into a mixture of 10 ml. of commercial formalin (40 per cent.) and 50 ml. of water acidified with 1 to 2 drops of conc. hydrochloric acid. If the resulting solution is slightly coloured by traces of heavy metals present in the formalin it should be filtered. The product should be kept at a low temperature in a closed vessel. It becomes turbid after 2 or 3 days. By certain modifications, described in the original, germanium can be detected in presence of 100 times its amount of arsenic, tin, selenium (as selenite) and other elements.

E. B. D.

Reduction of Molybdate in the Silver Reductor. C. F. Hiskey, V. F. Springer and V. W. Meloche. (*J. Amer. Chem. Soc.*, 1939, **61**, 3125–3127.)—In confirmation of the work of Birnbaum and Walden (*id.*, 1938, **60**, 64) it was found that when molybdate in 2 *N* hydrochloric acid solution is passed through the silver reductor and the reduced solution is exposed to the air the molybdenum is present entirely in the quinquevalent condition.

S. G. C.

Microchemical

Quantitative Micro-Determinations with the Use of Ordinary Analytical Balances. A. A. Benedetti-Pichler and R. A. Paulson. (*Mikrochem.*, 1939, **27**, 339–347.)—Ordinary student-type analytical balances are used for the work, but with 5-mg. substituted for 10-mg. riders. The precision of each balance is determined for a load of 10 g. on each pan, and the sensitivity is adjusted to 5 or 6 divisions of the pointer-scale per mg.; one balance of rather poor precision was among those tested. The balances can be used, in conjunction with the Emich filter-stick technique, for the determination of different inorganic ions, *e.g.*,

sodium in sodium oxalate. The sample (6–10 mg.) is weighed into a platinum crucible, converted into sulphate and weighed as such. Percentages of sodium varying from 34.15–34.46, as compared with calculated value 34.38, were obtained, the average deviation from the mean being ± 0.08 per cent. With the poor balance the same deviation from the mean (5 determinations) was obtained. The balances have also been used for the determination of aluminium in alum by the 8-hydroxyquinoline method. With samples weighing 6 to 8 mg. the average deviation from the mean (a_s) was ± 0.04 per cent. of Al_2O_3 and the relative average deviation of a single determination from the mean (a_s') was 0.37 per cent.; with samples of 16 to 18 mg., $a_s = \pm 0.03$ per cent. of Al_2O_3 and $a_s' = \pm 0.28$ per cent. In determinations of calcium in Iceland spar by the oxalate method on samples of 16 to 18 mg., $a_s = \pm 0.20$ per cent. of CaO and $a_s' = \pm 0.36$ per cent. In the determination of magnesium by the ammonium phosphate method $a_s = \pm 0.07$ per cent. of MgO and $a_s' = \pm 0.41$ per cent. Micro-methods can therefore be put into general practice without expenditure on a micro-balance. J. W. M.

Salts of Complex Cations Applied to the Microscopical Detection of Anions. W. A. Hynes and L. K. Yanowski. (*Mikrochem.*, 1939, 27, 336–338).—*Mono-aquopentamminocobaltic chloride* (roseocobaltic chloride).—The reagent is added as a solid of the size of a pinhead, at one side of the test drop containing about 1 per cent. of the ion to be tested. Characteristic crystals are obtained with the following substances:—oxalates, sulphasalicylic acid, dithionates, ferri- and ferrocyanides, silicofluorides and phosphomolybdates. Of these, only the first four reaction products are recommended for identification of the corresponding anions. No reactions are obtained with benzoic, boric, citric, formic, maleic, malonic, oxalic, salicylic, succinic, sulphanilic or uric acid, or with the alkali metal salts of the following anions:—acetate, azide, bicarbonate, bisulphate, bisulphite, bitartrate, borotartrate, bromate, bromide, carbonate, chlorate, citrate, dichromate, fluoride, formate, hypophosphite, iodide, metaborate, nitrate, nitrite, nitroprusside, orthoarsenate, orthoarsenite, secondary orthophosphate, perchlorate, permanganate, pyrophosphate, selenate, selenite, sulphate, tartrate, tellurite or thiocyanate. Eight photomicrographs are given. J. W. M.

Chemical Microscopy of Some Toxicological Alkaloids. W. F. Whitmore and C. A. Wood. (*Mikrochem.*, 1939, 27, 249–334).—The behaviour of each of twenty alkaloids with different types of reagents was investigated. The alkaloids include the following:—*local anaesthetics*: β -eucaine, cocaine, procaine, stovaine; *cinchona alkaloids*: cinchonine, cinchonidine, quinine, quinidine; *opium alkaloids*: codeine, dionine, heroine, morphine, narcotine, papaverine; *nux vomica alkaloids*: brucine, strychnine; *purine bases*: caffeine, theobromine; *miscellaneous*: atropine, nicotine. Optical properties of the crystalline products, with the exception of refractive indices, were ascertained with the aid of a polarising microscope. The reagents included the following:—I *Halogen reagents* (1) *Bromine in sodium bromide solution*.—This is a general precipitant for all alkaloids; it forms crystalline precipitates with six alkaloids, the best with theobromine, caffeine and atropine. (2) *Iodine trichloride* gives precipitates of little value for identification purposes. II *Oxygen acids and salts, including organic salts*.—(3) (a) *Perchloric acid* gives

distinctive derivatives with strychnine, brucine, morphine and cocaine; (b) *ammonium hypochlorite* forms crystals also with novocaine, cinchonine and quinine. (4) *Sodium perborate* yields the free bases. (5) *Ammonium vanadate* forms crystals with strychnine but they are not very characteristic. (6) *Potassium anthraquinone sulphonate*.—The best crystals are obtained with quinidine. (7) *Potassium xanthate*.—Only brucine and strychnine give crystalline xanthates. (8) *Cupferron*, crystals not well developed with β -eucaine. (9) *Potassium acid phthalate* forms crystals with strychnine. (10) *Potassium bromide* gives crystals with β -eucaine, codeine and stovaine. III *Salts leading to complex or double salt formation*.—(11) (a) *Stannous chloride* gives crystals with strychnine, brucine, β -eucaine and cocaine; (b) *Stannous bromide* behaves like (a) but forms better crystals with cocaine. (12) *Titanous chloride* gives characteristic crystals with strychnine, brucine and papaverine. (13) *Bismuth trichloride* forms crystals with strychnine and brucine. (14) *Potassium antimonyl tartrate*.—Strychnine gives isotropic crystals. (15) *Arsenic trichloride* does not form crystalline precipitates. (16) *Mercurochrome* forms non-crystalline precipitates. IV *Anions of complex salts or acids*. (17) *Sodium silver iodide* gives a characteristic crystalline iodoargentate precipitate with cocaine. (18) (a) *Potassium auro-iodide* is a general precipitant; caffeine and nicotine form crystalline products; (b) *Hydrogen auro-iodide* (HAuI_2) behaves like (a) except that a crystalline precipitate is obtained with theobromine. (19) (a) *Sodium auro-bromide* (NaAuBr_4) and (b) *Hydrogen auro-bromide* (HAuBr_4) give more sensitive tests than the chloro compound. (20) *Iridium chloride* (IrCl_3) behaves like (24) but the crystals are more soluble. (21) and (22) *Rhodium chloride* (RhCl_3) and *Ruthenium chloride* give crystals with strychnine and brucine, and ruthenium chloride also produces, with cocaine, a crystalline precipitate with oblique extinction. (23) *Sodium osmium chloride* (NaOsCl_6) behaves like (20), forming yellow precipitates. (24) (a), (b) and (c) *Hydrogen platinumchloride* (H_2PtCl_6) in 20 per cent. HBr, and in 5 per cent. HBr and *Sodium platinibromide* (Na_2PtBr_6). These reagents are more sensitive than the generally used chloroplatinic acid. Good crystals are obtained with dionine and with cocaine, for which the test is highly selective; cinchonine crystals show oblique extinction. (25) *Potassium platonic iodide* (K_2PtI_6) is a sensitive reagent giving with all the alkaloids precipitates of which the cinchonine compound is the most characteristic. (26) *Caesium cadmium iodide* (Cs_2CdI_4) is similar to (27) (a), *cadmium iodide* (CdI_2), which forms characteristic crystals showing oblique extinction with codeine, parallel extinction with morphine; (b) *Cadmium bromide* is similar to (a), but the crystals with novocaine are characteristic; (c) *Cadmium chloride* gives soluble crystals with heroine. (28) (a) *Potassium mercury bromide* (K_2HgBr_4) and (b) *Potassium hydrogen bromide* (K_2HgBr_4), when acidified, give insoluble salts with all the alkaloids; (a) gives crystals with morphine, nicotine, codeine and brucine, (b) also with strychnine, dionine and β -eucaine. (29) *Potassium mercury thiocyanate* ($\text{K}_2\text{Hg}(\text{SCN})_4$) is useful as a general precipitant. (30) *Potassium thallic iodide* gives only amorphous precipitates. (31) (a) *Hydrogen stannic chloride* (H_2SnCl_4) is a very useful reagent, yielding crystalline derivatives with those alkaloids that produce crystals with stannous chloride; these compounds have distinctive optical properties; (b) $\text{H}_2\text{SnCl}_4 + \text{KBr}$ forms crystals similar

to those obtained with (a) but smaller. (32) *Hydrogen antimony chloride* (HSbCl_4) is a good reagent for strychnine. (33) *Manganese sulphate + ammonium thiocyanate*. (34) *Cobalt nitrate + ammonium thiocyanate*. (35) *Chromium sulphate + ammonium thiocyanate*. (37) *Cadmium nitrate + ammonium thiocyanate*. Complex thiocyanates were obtained with nicotine and brucine. (38) (a) *Potassium palladium bromide* and (b) *Palladium chloride* in HBr give precipitates suitable for identification under the microscope with β -eucaine, brucine, strychnine and caffeine; the crystals, however, are not particularly distinctive. (39) *Sodium stannic iodide* (Na_2SnI_4).—The most distinctive crystals are obtained with cinchonidine. There are 132 drawings and photomicrographs of crystals. J. W. M.

Micro-Estimation of Threonine. R. J. Block and D. Bolling. (*J. Biol. Chem.*, 1939, **130**, 365–374.)—A practicable method of estimating small amounts of threonine (α -amino- β -hydroxybutyric acid) has been worked out, in which the amino-acid is quantitatively oxidised to acetaldehyde by means of lead tetra-acetate solution, and the acetaldehyde is then condensed with *p*-hydroxydiphenyl, with the formation of an intense red-violet colour (Eegriwe, *Z. anal. Chem.*, 1933, **95**, 323). The intensity of the colour is proportional to the amount of acetaldehyde and thus to the amount of threonine. The apparatus consists of 6 tubes connected together in series by means of rubber tubing. Tube 1 is a Pyrex test-tube (20×2.5 cm.) fitted with a rubber stopper carrying a long and a short tube, and containing 20 ml. of conc. sulphuric acid, which serves to clean and dry the air that is bubbled through it. Tube 2 is a blank tube exactly like tube 1, and serves as a trap for the sulphuric acid spray. Tube 3 is the reaction tube, and consists of a 20×3 cm. Pyrex test-tube with a standard ground-glass joint, into which fits a head containing an air inlet tube of 4 mm. inside diameter, extending to within 2.5 cm. from the bottom, and an air outlet. The tube is immersed in a water-bath maintained at 30°C . It contains the reaction mixture, consisting of 25 ml. of glacial acetic acid (purified by refluxing for 7 hours with 0.5 to 1.0 per cent. of potassium dichromate and then distilling), less than 10 mg. of amino-acids, and at least 1 g. of lead tetra-acetate (recrystallised from boiling glacial acetic acid). Tube 4 is similar to tube 1 and contains a 2-inch column of sodium hydroxide pellets to remove the last traces of acid; it is kept at room temperature. Tube 6 is the absorption tube and is of the same construction as tube 3. It contains 10 ml. of conc. sulphuric acid (analytical grade that has been allowed to stand over a small amount of lead shavings), 5 drops of water and 100 mg. of *p*-hydroxydiphenyl (purified by recrystallisation several times from acetone) in suspension; this tube is kept at 0°C . Air is drawn through the system at a constant rate for 1 hour. No pink or blue colour should be found in tube 6 when a blank estimation is carried out with the reagents only; an appreciable blank is usually due to impurities in the acetic acid. At the end of the reaction tube 6 is kept at 0°C . until it is desired to remove the excess of *p*-hydroxydiphenyl, when this tube is immediately transferred to a boiling water-bath for exactly 2 minutes (stop-watch). The tube is immediately cooled to 0°C . and subsequently allowed to attain room temperature. The colour, which varies from red to violet, depending on the amount of acetaldehyde, on the time, and on the temperature, is stable for 24 hours. The solution

absorbs strongly in the green part of the spectrum and has an absorption maximum at $560\text{m}\mu$. The colour can be measured in an Evelyn photoelectric colorimeter with filter $560\text{m}\mu$, a Duboscq visual colorimeter with the same filter, or a Pulfrich step-photometer with filter S57. A specimen of commercial synthetic *dl*-threonine is employed as standard, or a calibration curve may be constructed. All the other amino-acids tested failed to give colours, with the exception of *dl*-serine and *dl*-alanine, 10 mg. of which formed as much acetaldehyde as 0.03 mg. and 0.5 mg. of threonine respectively. A method of overcoming the interference of alanine is being investigated. All the carbohydrates tested yielded no detectable amounts of acetaldehyde on oxidation with lead tetra-acetate: Recoveries of 97 ± 3 per cent. of threonine added to protein hydrolysates were obtained. The following amounts of threonine were found in different kinds of protein:—casein, 3.5 per cent.; gelatin, 0.5 per cent.; normal human serum protein, 5.9 per cent. F. A. R.

Physical Methods, Apparatus, etc.

Determination of Water in Ether. R. Gaspart and G. Serrure. (*Bull. Soc. Chim. Belg.*, 1939, 48, 283–292.)—Gaspart's earlier work on the determination of water in acetone by means of measurements of absorption-spectra in the far infra-red (*cf.* ANALYST, 1939, 459) has been extended to solutions and suspensions in ether. Ether was purified by fractional distillation in presence of sodium metal in the dark (*cf.* Timmermans and Martin, *J. Chim. Phys.*, 1928, 25, 434) in a Pyrex glass apparatus, through which was passed a current of pure dry nitrogen. An apparatus for the accurate preparation of solutions of water of different concentrations in ether, and for the transference of these to the cell of the spectroscope without appreciable contamination from external sources, is described and illustrated. The method makes possible the preparation of solutions of known concentration, with an accuracy ranging from 0.5 to 14 per cent. according to the concentration. The cell was cooled in a mixture of solid carbon dioxide and alcohol during these operations, and at once hermetically sealed; measurements were carried out with a thickness of liquid of 1 mm. and at 20°C ., over the range 3700 to 3400 cm^{-1} (*cf.* ANALYST, *loc. cit.*) on solutions containing 0 to 50 parts of water per 100,000 by volume. The data are tabulated, and curves are plotted showing the relationship in each solution between the wave-length and the percentage transmission as obtained from the ratio of the energy-equivalent of the radiation after traversing the solution in question, to that obtained after passage through a comparison cell containing carbon tetrachloride. It is considered that the error of measurement, which arises mainly from reflection at the air-cell and cell-liquid interfaces, is of the order of 0.5 per cent. The curves show that for low concentrations of water a band (due to water-ether addition compounds) is obtained at 3598 cm^{-1} . This band increases in magnitude as the concentration increases, until at 6.6 p.p. 100,000 a second band (at 3525 cm^{-1}) becomes apparent. With acetone (*loc. cit.*) this second band was masked by the band (at 3420 cm^{-1}) characteristic of the $-\text{C}:\text{O}$ linkage, the first band (due to the acetone-water addition compound) being at 3618 cm^{-1} . Curves showing the ratio of the above percentage transmission of each solution to that of pure ether as a function of the

wave-lengths, demonstrate these two bands clearly and show how their magnitude (and especially that of the first) is dependent on the concentration of the water. The absolute and molecular coefficients of extinction for the band 3598 cm.^{-1} are plotted against the concentration. The molecular coefficients are less than the corresponding values for acetone (*loc. cit.*), and since they are only constant for concentrations exceeding $1:21 \times 10^{-6}$, it is therefore necessary to construct a standardisation curve; the sensitiveness of the method then corresponds with a concentration of the order of $1:2 \times 10^{-6}$. Similar data and curves are given for colloidal solutions of gold in ether containing various quantities of water. J. G.

Reviews

INTRODUCTION TO PRACTICAL ORGANIC CHEMISTRY. By F. G. MANN and B. C. SAUNDERS. Pp. ix + 191. London, New York and Toronto: Longmans, Green & Co., Ltd. 1939. Price 4s. 6d. net.

This book is a "severely abridged version" of the authors' larger work—*Practical Organic Chemistry*. It is intended as an introduction to the practical side of this branch of chemistry. It differs from the authors' advanced book in that the more difficult estimations and compounds, and also the sections on vacuum distillation and enzyme action are omitted.

Part I is devoted to methods of manipulation. In it careful and detailed, if somewhat lengthy accounts, are given of most of the fundamental processes used for the purification of organic compounds. The criteria for ascertaining the success of these operations are also fully discussed. In the course of this discussion many interesting (and too often forgotten) facts are mentioned. Hints contributory to success and to the avoidance of accidents are given freely.

Part II is concerned with preparations; in all, about forty-five are described, the aliphatic and aromatic series being about equally represented. In many instances alternative methods are given. The chief reactions and points of theoretical interest connected with each compound are also set forth clearly. The section on sugars is especially good; all the common types seem to be represented.

The next section is taken up with the reactions and methods of identification of organic compounds. The scheme, which is on the orthodox lines, is satisfactory and comprehensive, covering most of the common classes of compounds. In testing for elements Middleton's zinc and sodium carbonate method is recommended. Timely warning is given of the dangers attached to the sodium method when used with polynitro compounds and with carbon tetrachloride or chloroform. The special reactions for the identification of individual compounds seem to have been selected with great care.

The final chapter deals with various quantitative analyses. Among those described are the determination of molecular weights and methods for the determination of nitrogen (Kjeldahl), aniline, urea and acetyl groups.

The book closes with an appendix containing, *inter alia*, notes on the preparation of reagents, on first aid and on fire. Logarithms, antilogarithms and a good index are provided.

An examination of the book will show that a very satisfactory course of introductory practical organic chemistry is given. Students who have worked faithfully through it will have gained a practical knowledge that will be invaluable, not only as a basis for further organic laboratory work, but also for a clear understanding and appreciation of the theoretical side of the subject.

HAROLD TOMS

AN INTRODUCTION TO CRYSTAL CHEMISTRY. By R. C. EVANS, M.A. Pp. xi + 388. Cambridge University Press, 1939. Price 18s.

The story of the development of crystal chemistry (and physics) is a remarkable instance of the dependence of a branch of science on the assistance furnished by the inventor and designer of instruments. True, certain regularities were apparent to the seeing eye, and were duly noticed, but the story of modern crystallography begins with the invention of the goniometer by Carangeot in 1780, and was enormously advanced by the invention of the reflecting goniometer by Wollaston in 1809.

Much was accomplished in crystal chemistry—in which the chief interest lies “in the interpretation of the observed crystal structures and in their correlation with physical and chemical properties”—during the nineteenth century; but the major discovery of the diffraction of X-rays by a crystal acting as a grating, has resulted in most astonishing advances in our knowledge of the intimate structure of solids, advances which it is becoming increasingly necessary to present in such a form that the chemist of to-day may be able to appreciate their significance and importance.

It is too commonly assumed that this appreciation demands an equipment surpassing that possessed by the chemist who has not had the advantage of an early and systematic training in the modern outlook and technique. This is, perhaps, true if attention is concentrated on what may be termed the topographical aspect of the subject, but Dr. Evans has demonstrated that it is possible to begin at the beginning and to give his readers a comprehensive picture of modern views on the crystal lattice, and of the applications of these views to such topics as metallic elements and alloy systems, homopolar compounds, and ionic and molecular compounds, without making heavy demands on their mathematical knowledge.

Dr. Evans's book may be unreservedly commended as an admirably clear account of a fascinating and important subject.

ALLAN FERGUSON

FAT, TOTAL SOLIDS AND MOISTURE. By R. D. MASON, M.Sc., A.I.C. Pp. 101. London: A. Harvey. 1939. Price 7s. 6d. net.

The standardisation of control tests is essential in modern industry, and by this is meant the use of standard apparatus as well as technique. In connection with the determination of total solids by drying *in vacuo* and the determination of fat by the well-known Rose-Gottlieb method, this standardisation is carried to a high level by the ingenious Mojonnier tester, now well known to most chemists. This very excellence, however, seems to have stultified any effort to overcome certain defects that it undoubtedly possesses. An English machine has now been made which has taken advantage of the criticisms levelled at the

original one, and this book gives in a concise, and therefore extremely useful, form the details for testing by its means a large range of food products for fat and total solids. The value of the book is enhanced by an appendix giving tabulated data for all the products dealt with. It is strongly recommended to those who can afford the unit.

E. B. ANDERSON

ESSENTIALS OF PHYSIOLOGICAL CHEMISTRY. By ARTHUR K. ANDERSON. Pp. 323 + ix. New York: John Wiley & Sons, Inc.; London: Chapman & Hall, Ltd. 1939. Price 13s. 6d.-

When the first edition of this book was reviewed just over three years ago (*cf.* ANALYST, 1936, 61, 443), it was suggested that a new edition would be needed "in the not very distant future," but the extent of the revision necessary could hardly have been foreseen at that time. Indeed, a comparison of the two editions gives one some idea of the vast amount of research work that has been carried out during this period, and emphasises the necessity of revising adequately, or perhaps one should say re-writing, textbooks of physiological chemistry, every few years. One hopes that the obvious success of "*Essentials of Physiological Chemistry*" will encourage Professor Anderson to revise just as thoroughly each future edition that may be required.

The volume under review contains thirty more pages than its predecessor, including an entirely new chapter on "The Composition of Tissues." This is an especially interesting section of the book, as it deals mainly with substances of high molecular weight, the composition of which can at present only be guessed at, and one visualises this chapter being expanded into several as the years go by and our knowledge of these substances increases.

Of the other chapters, there are four or five that have especially increased in size. These are the chapters on "Carbohydrates," with nine additional pages; on "Lipids," with seven extra pages; on "Fat Metabolism" and "Carbohydrate Metabolism," the latter, in particular, having been largely rewritten to conform with modern theory. As one might expect, the chapters on "Enzymes," "Endocrine Organs" and "Vitamins" have also had to be enlarged, the last chapter showing the biggest proportional increase of them all. New subjects dealt with in these chapters include the oxidases, co-enzymes, sex hormones, the hormones of the pituitary gland, the vitamin B complex and vitamin E.

One striking exception to the general tendency to devote more space to everything is provided by the account of the chemistry of muscle contraction, which is reduced from two pages to two paragraphs; one can only conclude from this that the author has grown less dogmatic on this subject as the years have gone by!

There are one or two errors to which attention should be directed; thus, "leucine" is spelt "lucine" on page 192, and Na_2CO_3 is surely intended for NaHCO_3 in the formula on p. 234, but alas, the besetting sin of the last edition, an apparent difficulty in presenting accurate structural formulae, is again in evidence in this edition. Thus the formula for vitamin B_1 is given on p. 287 without the bridge methylene group, and the formula given on p. 267 for corticosterone does not include the angle-methyl groups, and, in addition, an oxygen atom is shown in

ring C instead of a hydroxyl group. The formula of α -tocopherol on p. 301 would have been less ambiguous if the double bonds of the benzene ring had been indicated.

Whilst it is unfortunate that the accuracy of a textbook of this nature should be impaired by such minor errors as these, it should be clearly understood that the book is a reliable guide, well written and well produced, to that borderland of science that lies between biology and chemistry. One could, of course, if one were so minded, quarrel with several of Professor Anderson's more controversial statements, but no author could be expected to enter upon a full discussion of every question that is open to doubt in a textbook of this kind, and one can only hope that future editions will bear the same impress of wide reading and careful assimilation as does this edition.

F. A. ROBINSON

VITAMIN E. A Symposium held under the Auspices of the Food Group (Nutrition Panel) of the Society of Chemical Industry. April, 1939. Pp. viii + 88. Cambridge: W. Heffer & Sons, Ltd. Price 5s.

Every time a new war breaks out the names of unheard-of towns become familiar; in the same way each time another vitamin is isolated another obscure reagent claims a place in one's memory. On this occasion it is allophanic acid, since it is in the form of crystalline allophanates that vitamin E is isolated. The vitamin itself is now called tocopherol. The classically educated mind may puzzle for a moment to decide from whence these names are derived.

The volume under review, as its title implies, is made up of a number of addresses delivered at a symposium last spring, and it presents in a vivid manner the state of knowledge of vitamin E at that time. The first section of the book, dealing with the chemistry of the vitamin, begins with a clear-cut review by Professor A. R. Todd of the isolation of vitamin E, the elucidation of its structure, and finally its synthesis. The narration of elegant work by the organic chemist leading to the final success of a synthetic production of a natural substance is always fascinating, and every chemist will read this survey with pleasure. Other articles follow, dealing with synthetic analogues of the vitamin and methods for its chemical estimation. It appears that a promising method for the determination of vitamin E is based on its power to reduce ferric chloride.

The satisfaction induced in the reader by the tale of achievement recounted in the first part of the book begins to evaporate as soon as he enters the two sections that follow. The physiological functions of vitamin E and its usefulness in human and veterinary therapeutics are beset with doubts and contradictions. These doubts weigh increasingly on the reader as, having followed a review by Professor J. C. Drummond of the confusing physiological situation, he is plunged into the thick of the trouble in articles dealing with the difficulties of producing standard conditions of vitamin E depletion in rats, and hence the difficulty of measuring the vitamin. Other contributions are concerned with such points as the relation of vitamin E to the endocrines and the distribution of vitamin E in rat tissues.

Vitamin E has been used therapeutically in both veterinary and human medicine. An excellent review by P. Vogt-Möller covers the literature in both fields. Then comes a paper by E. Shute expounding his theory that vitamin E

and oestrogens are in antagonistic equilibrium in the blood. By this time the scientific reader begins to appreciate that he has left the realms of exact experimentation and entered the field of clinical research where the doctor who is reluctant to make negative controls of any of his patients is forced to draw his conclusions without them. The present practical position is summed up thus by Professor F. J. Browne, who was chairman of the clinical session of the symposium: while one worker claims that habitual abortion can be cured with vitamin E, another states that it cannot but that the vitamin will prevent threatened abortion. He is contradicted by a third. Where seven women out of eighteen cases of habitual abortion, however, *were* reserved as negative controls and were *not* given vitamin E, all gave birth normally. Thus at present the published findings of the clinical use of vitamin E cancel themselves out.

The bibliography at the end of the book is one of its most valuable parts and will recommend it to all those working on the subject. For the more general reader the volume provides at once a clear summary of the present state of knowledge of vitamin E written by many of the leading workers in the field; at the same time the presentation as a series of papers gives a vivid impression of the discoveries of the future in the making. Finally, while the Public Analyst values the establishment of a chemical method for the assay of vitamin E preparations, he awaits, with perhaps even more interest, the decision of the biologist as to whether the human species is allied with the rat, which requires vitamin E for fertility, or claims relationship to the goat, which multiplies happily without it.

MAGNUS PYKE

Publications Received

BELL'S SALE OF FOOD AND DRUGS. 10th Ed. By R. A. ROBINSON and R. IVES. Pp. xxviii + 363. London: Butterworth & Co. (Publishers), Ltd. 1939. Price 15s. net.

FLUORESCENCE ANALYSIS. By J. A. RADLEY and J. GRANT. 3rd Ed. Pp. xvi + 424. London: Chapman & Hall. 1939. Price 22s. 6d. net.

MICRO-DIFFUSION ANALYSIS AND VOLUMETRIC ERROR. By E. J. CONWAY. Pp. xiii + 306. London: Crosby Lockwood & Son, Ltd. 1939. Price 25s. net.

METALLURGICAL ANALYSIS AND ASSAYING. By J. S. REMINGTON and F. L. JAMESON. Pp. 101. London: The Technical Press, Ltd. Price 5s. net.

TEXT-BOOK OF QUANTITATIVE CHEMICAL ANALYSIS. Y. H. YOE. Pp. ix + 219. New York: Wiley. London: Chapman & Hall. 1939. Price 12s. 6d. net.

BRITISH CHEMICALS AND THEIR MANUFACTURERS. Association of British Chemical Manufacturers. Pp. 393. 1939.

REPORT OF THE DEPARTMENT OF HEALTH OF MONTREAL (CANADA) FOR THE YEAR 1938.



THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

Deaths

WITH deep regret we record the deaths of the following members:

Mr. Frank George Edmed, a former Member of Council (January 22nd).

Sir Gilbert Morgan, Honorary Member (February 1st).

Dr. Frank T. Shutt, Formerly Dominion Chemist, Ottawa (January 5th).

The Phytic Acid Content of some Poultry Feeding Stuffs

By R. H. COMMON, PH.D., A.I.C.

IN view of the probable significance of phytic acid in the calcium-phosphorus metabolism of the fowl (Lowe, Steenbock and Krieger,¹ Common²) the phytic acid phosphorus content of a number of feeding stuffs as sold for poultry feeding has been determined in this laboratory. The method adopted was essentially that of McCance and Widdowson,³ modified with a view to convenience when large numbers of analyses are being made simultaneously.

A quantity of the feeding stuff containing about 4 to 8 mg. of phytic acid phosphorus was dried at 100° C. and extracted for three hours with 100 ml. of $N/2$ hydrochloric acid in an end-over-end shaker. The extract was filtered, and 25 ml. of the filtrate were pipetted into a 50-ml. graduated flask, neutralised to phenolphthalein with 25 per cent. sodium hydroxide solution, re-acidified with a few drops of $N/2$ hydrochloric acid, made up to the mark and mixed. Aliquot portions (usually 20 ml.) for duplicate determinations were pipetted into 50-ml. centrifuge tubes and made up to 20 ml. with water when necessary. Four ml. of a solution of ferric chloride in N hydrochloric acid, containing 1.0 g. of iron per litre, were then introduced into each tube, and the tubes were heated in the water-bath for fifteen minutes. Each tube was then cooled and centrifuged, the supernatant liquid was decanted from the precipitate of ferric phytate, and the tube was drained and dried inside with filter-paper. The precipitate was broken up with 2 to 4 ml. of $N/2$ hydrochloric acid and re-centrifuged, the wash liquid being poured off and the inside of the tube drained and dried as before.

The precipitate was broken up by blowing in 2 ml. of water from a fine pipette, and decomposed by heating for fifteen minutes on the water-bath after further addition of 2 ml. of 2 per cent. sodium hydroxide solution. The contents of the tube were filtered through a 7-cm. filter-paper (Whatman No. 40) into a 50-ml. silica dish, hot water being used for washing. Two ml. of 20 per cent. w/v calcium acetate solution were then mixed with the filtrate, the mixture was dried at 100° C. overnight in the electric oven, and the residue was gently ignited in an electric muffle to a white ash. This was taken up by heating with about 6 ml. of 2 *N* hydrochloric acid and washed with hot water through a filter into a 100-ml. graduated flask. The contents of the flask were nearly neutralised with 25 per cent. sodium hydroxide solution and cooled, and phosphorus was determined directly by the method* of Fiske and Subbarow.⁴

From time to time the silica dishes used in the determination should be cleaned with hydrofluoric acid.

TABLE I

COMPARISON OF PERCHLORIC ACID OXIDATION AND DRY ASHING
TECHNIQUES IN PHYTIC ACID PHOSPHORUS DETERMINATIONS

| | | Phytic acid phosphorus | |
|------------|-------|--|---|
| | | By wet ashing with sulphuric and perchloric acids Per Cent. | By dry ashing with calcium acetate Per Cent. |
| Wheat meal | .. | 0.264 | 0.262 |
| | | 0.256 | 0.262 |
| Maize meal | .. | 0.192 | 0.257 |
| | | 0.213 | 0.259 |
| Pollard | | 0.194 | 0.250 |
| | | 0.209 | 0.254 |

The modified method was tested on a sample of pure sodium phytate prepared by Professor D. C. Harrison. The total phosphorus of the sample was found to be 10.8 per cent. and the phytic acid phosphorus to be 10.7 per cent., so that the method is capable of giving a recovery of phytic acid phosphorus of the order of 98 to 99 per cent.

* FISKE AND SUBBARROW'S COLORIMETRIC METHOD.—The following reagents are required:—
(a) *Molybdate solutions*.—A solution of 25 g. of ammonium molybdate in 400 ml. of water is added to 500 ml. of 10*N* sulphuric acid and made up to 1 litre with water. (b) *1:2:4-aminonaphthol sulphonic acid reagent*.—0.5 g. of the acid, 30 g. of sodium bisulphite and 6 g. of sodium sulphite (crystals) in 250 ml. of water. The standard phosphate solution is prepared by dissolving 0.3509 g. of potassium dihydrogen phosphate in water, adding 10 ml. of 10 *N* sulphuric acid and making up to 1 litre (5 ml. contain 0.4 mg. of phosphorus).

The aliquot part of the phosphate solution under examination, which should contain about 0.4 mg. of phosphorus, and be only slightly acid, is diluted to about 75 ml., treated first with 10 ml. of reagent (a) and then with 4 ml. of reagent (b), shaken and made up to 100 ml. The final acid concentration is about 0.5 *N*.

The standard for comparison is prepared by mixing 5 ml. of the original standard phosphate solution, 70 ml. of water and 10 ml. of the molybdate reagent (a), adding 4 ml. of reagent (b) and making up to 100 ml. The colorimetric comparison is made after about 30 minutes. If the aliquot portion contains up to 1.0 mg. of phosphorus the comparison is made against 10 ml. of standard phosphate solution.

The modified method was also tested against the original method of oxidation with sulphuric and perchloric acids. In some instances agreement was good, but in others the perchloric acid oxidation seemed to give more erratic results, as may be seen from Table I. This may have been due to the difficulty of removing the last traces of perchloric acid; traces of perchloric acid have been shown to interfere with colour development in the method of Fiske and Subbarow (Snook).⁵

Experiments were carried out on the recovery of phytic acid phosphorus added to extracts of meals. The recovery was satisfactory, as may be seen from Table II.

TABLE II

RECOVERY OF ADDED PHYTIC ACID PHOSPHORUS FROM N/2
HYDROCHLORIC ACID EXTRACT OF WHEAT

| Solution | Phytic acid phosphorus found mg. |
|--|--|
| 20 ml. of N/2 HCl extract of wheat .. | 1.245 |
| 20 ml. of N/2 HCl extract of wheat + 5 ml. of sodium phytate solution | 1.760 |
| 5 ml. of sodium phytate solution .. | 0.525 |
| Recovery = $\frac{(1.760 - 1.245)}{0.525} \times 100 = 98$ per cent. | |

Total phosphorus was determined by a modification of the method of Fiske and Subbarow, which has been described elsewhere (Common⁶).

Table III gives maximum and minimum figures for the phytic acid phosphorus contents of samples of cereals and other feeding stuffs, and Table IV records the results obtained with typical samples of the different products. The origin of most of the samples was known.

TABLE III

| | Phytic acid phosphorus Per cent. on dry matter | | Total phosphorus Per cent. on dry matter | | Phytic acid phosphorus Per cent. of total phosphorus | |
|---------------------------------------|---|-------|--|-------|---|------|
| | Max. | Min. | Max. | Min. | Max. | Min. |
| Wheat (11 samples) .. | 0.305 | 0.111 | 0.418 | 0.204 | 77.6 | 52.9 |
| Oats (5 samples) .. | 0.312 | 0.218 | 0.505 | 0.365 | 74.3 | 59.1 |
| Barley (3 samples) .. | 0.217 | 0.178 | 0.369 | 0.328 | 59.2 | 54.3 |
| Yellow maize meal (3 samples) .. | 0.314 | 0.277 | 0.407 | 0.364 | 78.3 | 76.1 |
| Bran (8 samples) .. | 1.030 | 0.512 | 1.571 | 0.571 | 77.7 | 59.3 |
| Extracted soya bean (3 samples) .. | 0.388 | 0.341 | 0.671 | 0.644 | 53.9 | 52.9 |

Phytic acid phosphorus accounted for about two-thirds to three-quarters of the total phosphorus of wheat, oats, barley and maize. The samples of barley examined tended to have a lower proportion of their phosphorus in the form of phytic acid than did the samples of wheat. About the same ratio of phytic acid phosphorus to total phosphorus was found in bran as in pollard of grades

similar to "weatings." Both total phosphorus and phytic acid phosphorus were lower in the Australian wheats than in the British wheats, in agreement with the observations of Snook (1938).⁵ Rice bran meal, tapioca root flour and hempseed all had about one-third of their phosphorus in the form of phytic acid phosphorus. Earthnut cake, extracted soya bean meal, maple peas, beans, millet and sunflower seed had from one-half to three-quarters of their total phosphorus in the form of phytic acid phosphorus. Dried grass contained only a very small proportion of its total phosphorus as phytic acid, alfalfa meal containing a greater proportion.

TABLE IV

| Feeding stuff | Place of origin | Crude fibre Per cent. on dry matter | Phytic acid phosphorus Per cent. on dry matter | Total phosphorus Per cent. on dry matter | Phytic acid phosphorus Per cent. of total phosphorus |
|-----------------------|-----------------------|--|--|--|--|
| Wheat meal | — | — | 0.301 | 0.458 | 65.7 |
| Oatmeal | Ireland | 11.24 | 0.242 | 0.355 | 68.3 |
| Oats | Ireland (Co. Down) | — | 0.272 | 0.434 | 62.7 |
| Sussex ground oats .. | Ireland | — | 0.218 | 0.365 | 59.7 |
| Pinhead oatmeal .. | Ireland | — | 0.299 | 0.495 | 60.5 |
| Barley meal | — | 5.03 | 0.216 | 0.355 | 61.0 |
| Yellow maize | River Plate | — | 0.276 | 0.368 | 75.0 |
| " | N. America | — | 0.149 | 0.199 | 75.0 |
| Bran | Australia | 10.92 | 0.512 | 0.676 | 75.8 |
| " | Canada | 12.54 | 0.974 | 1.258 | 77.5 |
| " | Holland | 12.36 | 1.030 | 1.571 | 65.5 |
| Pollard | — | 10.43 | 0.277 | 0.350 | 79.1 |
| White sharps | — | 7.55 | 0.312 | 0.467 | 66.8 |
| Fine pollard | River Plate | 7.19 | 0.962 | 1.238 | 77.7 |
| Coarse pollard .. | River Plate | 10.53 | 0.760 | 1.044 | 72.8 |
| Middlings | Holland | 6.98 | 0.481 | 0.936 | 51.3 |
| Fine white sharps .. | — | 7.03 | 0.364 | 0.558 | 65.3 |
| Rice bran meal .. | (E. Indies?) | 6.66 | 0.577 | 1.558 | 37.0 |
| Tapioca root flour .. | — | 1.91 | 0.028 | 0.083 | 33.8 |
| Earthnut cake | — | — | 0.402 | 0.668 | 60.2 |
| Dried yeast | — | — | 0.058 | 1.514 | 3.8 |
| Alfalfa meal | Canada | — | 0.027 | 0.131 | 20.5 |
| Dried grass | Ireland | — | 0.007 | 0.351 | 2.0 |
| Millet | Smyrna | — | 0.206 | 0.289 | 71.2 |
| Sunflower seed | Hungaria | — | 0.374 | 0.453 | 82.8 |
| Hempseed | Manchuria | — | 0.285 | 0.692 | 41.2 |
| Beans | England | — | 0.501 | 0.662 | 75.7 |
| Maple peas | Tasmania | — | 0.194 | 0.334 | 58.1 |

Small amounts of phytic acid were found in dried yeast. This presumably came from the malt, since phytic acid has not been recorded among the phosphorus compounds present in yeast (Macfarlane⁷).

It is evident that in an ordinary poultry ration about three-quarters of the phosphorus derived from cereals will be in the form of phytic acid. Experiments designed to ascertain the minimum calcium and phosphorus requirements of poultry should take cognizance of this fact.

I wish to record my indebtedness to Mr. F. Dickinson, F.I.C., Chemical Research Division, Ministry of Agriculture for Northern Ireland, Belfast, who very kindly carried out the fibre determinations, and to those grain millers who furnished samples of known origin. I also wish to thank Professor D. C. Harrison, of Queen's University, Belfast, for his advice and helpful criticism.

REFERENCES

1. J. T. Lowe, H. Steenbock and C. H. Krieger, *Poultry Sci.*, 1939, **18**, 40.
2. R. H. Common, *Nature*, 1939, **143**, 379.
3. R. A. McCance and E. M. Widdowson, *Biochem. J.*, 1935, **30**, 252.
4. C. H. Fiske and Y. Subbarow, *J. Biol. Chem.*, 1925, **46**, 375.
5. L. G. Snook, *Empire J. Exper. Agr.*, 1938, **6**, 20.
6. R. H. Common, *J. Agr. Sci.*, 1936, **26**, 85.
7. M. G. Macfarlane, *Biochem. J.*, 1936, **30**, 1369.

AGRICULTURAL CHEMISTRY DEPARTMENT
THE QUEEN'S UNIVERSITY, BELFAST

The Precipitation of Aluminium Hydroxous Oxide* and its Solubility in Ammonia

By E. B. R. PRIDEAUX, M.A., D.Sc., F.I.C., AND J. R. HENNESS, B.Sc.

THE precipitation of alumina by ammonia and its residual solubility should be explicable in terms of the electrochemical properties of the hydroxide and by theories of the colloidal state, but the position is by no means clear. Also, experimental data as to the solubility under analytical conditions, as well as in pure ammonia solution, are very scanty.

Precipitation from sulphate solutions by means of alkalis follows a course which is determined by the amphoteric ionisations of the hydroxide, but is complicated by colloidal phenomena (Britton¹). Visible precipitation begins at about pH 4 but not until about pH 6.5 in chloride solutions.

This is attributed to the tendency of chlorides to form colloidal solutions, and the difference between the two salts may be accounted for by the accepted theories. Thus, aluminium chloride is hydrolysed in stages, the colloidal basic salt is then stabilised by aluminium ion and forms a positive micelle with a double layer of chlorine ions as figured in papers by Weiser *et al.*, Weiser and Gray.² If sulphate ion is present, it tends to discharge and precipitate the micelle as a basic salt. After precipitation is nearly complete, the pH rapidly increases, an inflection appears, at about pH 7, but this is *not* the isoelectric point of alumina itself, since the precipitate contains acid radicle. With further addition of alkali, the pH increases still further from 8 to 10.5, with solution of alumina as aluminate. In the precipitation by ammonia, which depends upon the almost complete hydrolysis of ammonium aluminate, well-known methods, referred to below, seem

* Since definite meanings are attached to the terms "hydroxide" and "hydrate," it seems that a more general term would be useful to cover these as well as the amorphous and colloidal hydrated oxides of uncertain composition. The term "hydroxous oxide," however, as used by H. B. Weiser (*Inorganic Colloid Chemistry*, Vol. II) refers only to those amorphous colloidal substances which are neither definite hydroxides nor definite crystal hydrates. Thus, under "hydroxous oxides" are classified those of iron, aluminium and chromium, *i.e.* in their most usual form, and without reference to the possibility (realised for iron) of preparing definite crystal hydrates by special methods.

to aim at the isoelectric condition. Now, although the dissociation constants of alumina are not accurately known, there is no doubt that the basic is considerably greater than the acidic (ca. 1×10^{-10}); therefore the calculated isoelectric point should be definitely higher than pH 7. Regulation of the hydrogen ion concentration by the ammonium chloride and ammonia mixture proceeds according to the usual equation, the maximum buffer effect being at pH 4.64 or, if $pK_w = 14.14$, at pH 9.5, and for any given mixture, $H^- = (NH_4Cl/NH_3) \times 10^{-9.5}$. The calculated values of pH have been checked by us in a Lovibond comparator, using phenol red.

Ratio:

| $\frac{\text{ammonium chloride}}{\text{ammonia}}$ | | 8 | 16 | 32 | 64 | 128 | 256 | 512 |
|---|----|-----|-----|-----|-----|-----|-----|-----|
| pH calculated | .. | 8.6 | 8.3 | 8.0 | 7.7 | 7.4 | 7.1 | 6.8 |
| pH observed | .. | 8.5 | 8.2 | 7.9 | 7.5 | 7.1 | 6.9 | 6.7 |

Since it is the practice to add considerable quantities of ammonium salt and only the slightest excess of ammonia, it is clear that the pH will never rise above 9.0 and may easily pass to the acid side of neutrality in hot solutions.

Some experiments designed to correlate the alumina left in solution with the pH values were made, in 1916, by Blum,³ who added ammonia to 200 ml. containing aluminium equivalent to 0.1 g. of Al_2O_3 and 5 g. of ammonium chloride up to the change-points of various indicators:

| | | <i>p</i> -Nitrophenol | Methyl red | Rosolic acid | Phenolphthalein |
|-------------|--|-----------------------|------------|--------------|-----------------|
| pH | | 6 | 6.5 | 7.5 | 9 |
| Al_2O_3 | | | | | |
| in filtrate | | 0.001 to 0.0012 | 0.0001 | 0.0001 to 2 | 0.0004 |
| g. | | | | | |

Hence it was concluded that dilute ammonia solution should be added to the nearly boiling solution until this was nearly alkaline to methyl red; it should then be boiled for not more than 2 minutes, and filtered at once. Further quantities of 0.5 to 0.2 mg. appear in the washings with 75 ml. of hot water, and 0.3 mg. in those made with 2 per cent. ammonium chloride solution. It is evidently difficult to establish pH conditions of minimum solubility by means of indicators, since the solution is poorly buffered at pH 6.5 to 7, and the solubility rises on the acid as well as on the alkaline side. The precipitate also coagulates poorly until a definite excess of ammonia is present. The small weights recorded might be affected by various errors. In the present investigation the solubilities under analytical conditions, as well as those of pure alumina, have been determined by one of the new colorimetric methods.

EXPERIMENTAL.—A solution of potash alum was made up to contain 0.9 g. of aluminium per litre, and 10 ml. of this solution together with 5 ml. of 4*N* ammonium chloride solution were taken for each experiment (*i.e.* 9 mg. of aluminium). In experiments 1 to 7, these 15 ml. of solution were heated to boiling in a beaker and 5 or 10 ml. of ammonia of various concentrations were added. In experiments 8 and 9, the ammonia solution was heated, and the alum and ammonium chloride solutions were added. In every instance the solution was quickly heated to boiling and filtered through a Whatman's No. 1 filter, the

operation lasting about 1 minute. The filtrate was boiled to remove excess of ammonia and frequently became turbid owing to decomposition of the alumina sol. The volume of solution remaining was then measured, and the amount of aluminium contained in each ml. was determined by the colorimetric method. B.D.H. spot test reagent, aurine tricarboxylic acid (0.4 g.), was dissolved in water, and excess of ammonia was added; it was then boiled to remove the excess of ammonia and made up to 200 ml., thus giving a 0.2 per cent. reagent. A standard solution of aluminium, containing 0.02 mg. per ml., was made from potash alum. The colorimetric determinations were made on standard and sample as described in the B.D.H. "*Reagents for Delicate Analysis including Spot Tests.*" Solutions containing more than 0.5 mg. of aluminium per ml. gave a red precipitate, and concentrations were therefore kept below this and arranged to show nearly equal colours for standard and sample.

The results are presented in tabular form.

TABLE I

| Expt. | [NH ₃] before filtration Moles per litre | [NH ₄ Cl] | (H ⁺) × 10 ¹⁰ calc. | Total Al in filtrate mg. | Al per ml. mg. | Ratio Al/NH ₃ | Al × 10 ⁻⁴ (OH) |
|-------|--|----------------------|---|-----------------------------------|----------------------|-----------------------------|-------------------------------|
| 1 | 0.467 | 1.0 | 6.77 | 0.168 | 0.0084 | 1.8 | 0.056 |
| 2 | 0.748 | 0.8 | 3.38 | 0.23 | 0.0093 | 1.24 | 0.031 |
| 3 | 0.935 | 1.0 | 3.38 | 0.27 | 0.0135 | 1.44 | 0.0455 |
| 4 | 0.972 | 1.0 | 3.25 | 0.34 | 0.0168 | 1.74 | 0.0545 |
| 5 | 1.556 | 0.8 | 1.625 | 0.46 | 0.0183 | 1.18 | 0.030 |
| 6 | 1.945 | 1.0 | 1.625 | 0.16 | 0.0082 | 0.42 | 0.0135 |
| 7 | 3.11 | 0.8 | 0.81 | 0.39 | 0.0156 | 0.50 | 0.0125 |
| 8 | 0.972 | 1.0 | 3.25 | 0.20 | 0.0101 | 0.20 | — |
| 9 | 1.556 | 0.8 | 1.63 | 0.65 | 0.026 | 0.40 | — |
| 10 | ammonia (0.880) about 18.3 | | | — | 0.198 | 1.28 | — |

The amount of alumina in solution increases with the ammonia, although not in strict proportionality, up to about 1.5 *N*. Above this concentration the amounts dissolved fall and tend to become irregular. The ratio of aluminium to ammonia is about the same in the strongest as it is in 0.75 *N* ammonia. The aluminium dissolved varies also very roughly with the hydroxyl ion concentration, since this, at nearly constant ammonium ion concentration is, of course, proportional to the free ammonia. The variability of these results is due not to experimental error, nor even principally to variable losses of ammonia from hot solutions, but much more to the fact that alumina, precipitated in this manner, is the most colloidal of all the forms, whose properties vary with the speed of coagulation, as is emphasised in "*The Theory of Quantitative Analysis*," by H. Bassett. This author considers that the soluble part is not an ammonium aluminate, but a colloidal alumina peptised by the ammonia. If the concentrations of dissolved alumina given in column 6 of Table I are present in 100 ml. from which a normal amount, say, 0.2 g. of alumina, has been precipitated, then the percentage losses in 0.5 to 1.5 *N* ammonia vary from 0.4 to 0.9. The total losses (column 5) in the 20 to 25 ml. of filtrate of course form a much larger proportion of the possible weight of the precipitate, corresponding to 9 mg. of aluminium.

As shown later, these losses are mainly due to the hydrous oxide in a state of supersaturation or peptisation.

SOLUBILITIES OF ALUMINIUM HYDROXIDE IN AMMONIA.—A sample of the purest sheet aluminium, supplied by the courtesy of Mr. A. N. D. Pullen and the British Aluminium Company, had a guaranteed purity of 99.995 per cent. Pieces of uniform size and shape were amalgamated with mercury in excess, and placed in ammonia solutions of different concentrations. In the first series the ammonia solutions were kept in open tubes; in the second, redistilled ammonia solutions were allowed to react with the amalgam in closed tubes. In some of the experiments colorimetric determinations of alumina were checked by weight.

Any threads of precipitated alumina were, of course, removed by filtration before the determination of solubility.

The following results give the maximum initial solubilities in ammonia and in complete absence of ammonium salts:

| | | | | |
|-------------------------------------|------|------|------|-------|
| Ammonia, Normality.. .. . | 5.0 | 0.75 | 0.50 | 0.25 |
| Aluminium mg. per ml. (after 1 day) | 1.08 | 0.36 | 0.12 | 0.125 |

Concentrations of alumina were maintained for several days when ammonia solutions were in contact with precipitates. The filtrates, however, began to deposit alumina, and the concentrations progressively decreased, and more rapidly in those solutions having the lower concentrations, as follows:

Filtrate from 5 *N* ammonia, 1.3 mg. of aluminium per ml.

| | | | |
|-----------------------------|-----|------|------|
| Time (days) | 1 | 2 | 3 |
| Aluminium mg. per ml. | 0.8 | 0.45 | 0.25 |

Filtrate from 0.75 *N* ammonia, 0.36 mg. of aluminium per ml.

| | | | |
|------------------------------|---------------|----------------|----------------|
| Time (days) | $\frac{1}{2}$ | $1\frac{1}{2}$ | $2\frac{1}{2}$ |
| Aluminium, mg. per ml. | 0.1 | 0.04 | 0.022 |

In the next series the effect of standing was investigated, when the concentration of ammonia was maintained, in contact with the precipitates. Pure redistilled ammonia interacted with aluminium amalgam in closed tubes furnished with Bunsen valves.

Concentration of ammonia, initial 5.84 *N*; final 5.37 *N*.

| | | | | |
|------------------------------|-----|-----|------|------|
| Time (days) | 1 | 2 | 5 | 6 |
| Aluminium, mg. per ml. | 1.1 | 1.1 | 0.18 | 0.10 |

Concentration of ammonia, initial 2.92 *N*; final 2.756 *N*.

| | | | | |
|------------------------------|-----|-----|------|-------|
| Time (days) | 1 | 2 | 5 | 6 |
| Aluminium, mg. per ml. | 1.2 | 1.2 | 0.54 | 0.125 |

DISCUSSION OF RESULTS.—When the metal dissolves with evolution of hydrogen in solutions that are well on the alkaline side of the isoelectric point it must be supposed that ammonium aluminate is formed at first. This can then decompose in the following ways:

(a) *By Loss of Ammonia.*—In the tubes that were left open it was noticed that the precipitates formed a very coherent skin on the surface.

(b) *By Hydrolysis of Ammonium Aluminate.*—This produces colloidal alumina which gradually gives a turbidity throughout the solution. In the experiments

with closed tubes precipitation is delayed, either because hydrolysis is diminished or because the resulting hydroxide is peptised by ammonia. The precipitate finally appears in thread-like forms, the presence of which does eventually lower the solubility, owing to the continued solution of the metal. Thus in 0.75 *N* ammonia the solubility after 2½ days is 0.022 mg., which is only about twice that in 0.75 *N* ammonia with much ammonium chloride. Although the initial solubilities in ammonia alone are 10 to 100 times those in presence of ammonium chloride, the final solubilities may be hardly greater. The effect of the salt, then, is principally to accelerate the formation of nuclei. These results also give a reason for the observation of Blum, that the presence of a small excess of ammonia is useful in aiding the coagulation of the precipitate. For this is then formed by way of ammonium aluminate, and not by way of the very gelatinous form obtained on first precipitation. The amalgam experiments show in every instance a precipitate which is dense and easily separated by filtration.

A procedure based on these facts is suggested. This is to add *N* to 2*N* ammonia to the solution containing the usual ammonium salt, then to digest the precipitate in a closed tube in the water-bath, and finally to remove the excess of ammonia and filter. It is hoped that further experiments will be made in these directions as opportunity offers.

REFERENCES

1. H. T. S. Britton, *J. Chem. Soc.*, 1925, **127**, 2121.
2. H. B. Weiser and G. R. Gray, *J. Phys. Chem.*, 1932, **36**, 2178.
3. W. Blum, *J. Amer. Chem. Soc.*, 1916, **38**, 1282.

UNIVERSITY COLLEGE
NOTTINGHAM

November 20th, 1939

The Analysis of Cadmium-plating Solutions

By G. STANLEY SMITH, B.Sc., A.I.C.

CADMIUM is usually deposited from a cyanide solution which contains cadmium and potassium cyanides and caustic alkali. Baths that have been in use for a considerable period contain also large quantities of ferrocyanide and possibly copper.

The analysis of used solutions presents certain difficulties. The cyanide in the form of the comparatively stable ferrocyanide does not assist in the formation of a good deposit and should not be included in the figure for alkali or cadmium cyanide, yet a distillation with acid to obtain the cyanide in a form suitable for titration with silver nitrate yields in addition at least half the cyanide of the ferrocyanide. Methods involving distillation with excess of sodium bicarbonate or the passage of carbon dioxide through the hot solution have been found to give high results, for ferrocyanide is unstable in the presence of even such a weak acid as carbonic acid. Again, direct electrolysis of the solution to determine the total cadmium often yields a very poor, non-adherent deposit containing iron and takes many hours before the electrode ceases to gain in weight.

For determining total cyanide, excluding ferrocyanide, successful use has been made of the observation of Feld¹ that lead cyanide and magnesium cyanide are easily decomposed on boiling to give hydrocyanic acid and the hydroxides of the metals, whilst the ferrocyanides are stable. For cadmium, copper and iron, the original solution is decomposed with sulphuric acid, the cadmium and copper are deposited together electrolytically in a dilute sulphuric acid medium, the solution of the deposit in nitric acid is electrolysed for copper, and iron is precipitated as hydroxide in the former electrolyte. The details are as follows:

CYANIDE, EXCLUDING FERROCYANIDE.—Place 5 ml. of the solution in a 500-ml. flask fitted with a rubber bung carrying a tap-funnel and a trap connected with an upright water-condenser dipping into a flask or bottle containing 10 ml. of 20 per cent. sodium hydroxide solution and sufficient water to cover the outlet of the condenser to a depth of at least half-an-inch. Introduce into the flask 150 ml. of water and 50 ml. of 0.5 *M* solution of lead nitrate. (Lead nitrate is to be preferred to a magnesium salt, because with the latter a very slight decomposition of ferrocyanide is liable to occur.) Lead hydroxide and carbonate are precipitated, and the amount of lead nitrate added should be sufficient to destroy all the caustic alkali and carbonate. Distil the solution rapidly down to a small bulk and titrate the distillate with 0.1 *N* silver nitrate solution, using a little potassium iodide as indicator. If the distillation has been carried sufficiently far, no more cyanide should be evolved when a further 100 ml. of water is placed in the flask and the distillation continued. Even when the contents of the flask have been allowed to become almost dry, no decomposition of ferrocyanide has been detected.

The number of ml. of silver nitrate required, divided by 6, gives the cyanide-content, calculated as CN, in ounces per gallon.

CADMIUM, COPPER AND IRON.—To 5 ml. of the original solution, diluted to prevent undue frothing, add a slight excess of dilute sulphuric acid and boil down carefully until fumes appear, adding nitric acid to destroy organic matter. Cool, wash down with water and evaporate to fuming again. Take up the residue in water, neutralise with ammonia and add 10 ml. of dilute sulphuric acid (sp.gr. 1.2) per 100 ml. Add a few drops of hydrogen peroxide and electrolyse to deposit cadmium and copper, using platinum electrodes. The cathode may be coppered first, but it does not appear to be necessary. The electrolysis with a rotating electrode should be complete in half to three-quarters of an hour, but a test should be made to ensure that it is complete. Weigh the deposit, dissolve in nitric acid, dilute so that the solution contains about 10 ml. of dilute nitric acid (sp.gr. 1.2) per 100 ml. and electrolyse to deposit copper. Weigh and deduct the weight from that previously found to obtain the amount of cadmium.

CYANIDE.—Total alkali cyanide and "free cyanide" are obtained by calculation as follows:—*Total cyanide.*—From the figure for cyanide given by distillation deduct the equivalent of the cadmium and copper obtained from the formulae $\text{Cd}(\text{CN})_2$ and CuCN and calculate to KCN. *Free cyanide.*—Since this is usually taken to be potassium cyanide in excess of that required to form a compound of the formula $\text{K}_2\text{Cd}(\text{CN})_4$, a corresponding deduction from the total cyanide should give the free cyanide. It should be recognised, however, that any figure for free cyanide is somewhat arbitrary.²

RESULTS.—In Table I solution A represents a plating solution containing 8.5 g. of cadmium per litre, excess of potassium cyanide and no ferrocyanide; solution B represents a solution containing 11.3 g. of cadmium per litre and ferrocyanide, when expressed as potassium cyanide, equivalent to 3.95 ml. of 0.1 *N* silver nitrate per 5 ml. of solution, the volume taken for distillation; the cuprous cyanide solution contained 6.4 g. of copper per litre, and was obtained by dissolving cuprous cyanide in excess of potassium cyanide.

Distillation with sulphuric acid or, in presence of ferrocyanide, with sulphuric acid and cuprous chloride (Williams¹), was used to obtain comparative figures for the titration of total cyanide, but in the case of solution B the result has been corrected for the quantity of ferrocyanide present and decomposed.

TABLE I

| Solution distilled | Method of decomposition | Silver nitrate (0.1 <i>N</i>) required ml. |
|---|--|---|
| 25 ml. of solution A | Dilute sulphuric acid | 39.85 |
| " " | Magnesium chloride | 39.65 |
| " " | Lead nitrate | 39.8 |
| 25 ml. of <i>M</i> /30 potassium ferrocyanide | Magnesium chloride | 0.4 |
| " " | Lead nitrate | nil |
| 25 ml. of each of the solutions above | Lead nitrate | 39.9 |
| 5 ml. of solution B | Lead nitrate | 17.9 |
| " " | Dilute sulphuric acid and cuprous chloride | 17.85 |
| 25 ml. of cuprous cyanide solution | Dilute sulphuric acid | 37.5 |
| " " | Magnesium chloride | 12.6 |
| | | (plus 1.35 on second distillation) |
| " " | Lead nitrate | 36.0 |
| | | (plus 1.55 on second distillation) |

A more rapid method for evaluating cadmium solutions would be useful, and it was thought that titrations with acid after adding formaldehyde to one portion and mercuric chloride to another might give (a) total cyanide, (b) cadmium, (c) hydroxide with carbonate, by calculation from these simple titrations. The method failed with the plating solutions used owing to the presence of ferrocyanide, but the principles are outlined in the event of someone finding it possible to adapt them.

On addition of formaldehyde, cadmium gives a precipitate of cadmium hydroxide, whilst potassium cyanide gives the potassium salt of glycollic nitrile, $\text{CH}_2(\text{OK})\text{CN}$, which is soluble and reacts alkaline. After filtration, titration of the filtrate, or preferably, for speed, an aliquot portion, should give a figure for alkali hydroxide and carbonate plus alkali cyanide.

With mercuric chloride solution, containing sodium chloride to repress hydrolysis, cyanides form mercuric cyanide, which is neutral. A determination

of the alkalinity gives that due to alkali hydroxide and carbonate. After neutralising and adding an excess of potassium iodide, which forms a more stable mercury salt and sets free potassium cyanide equivalent to the original potassium cyanide plus cadmium cyanide, a further titration with acid gives the total cyanide.

REFERENCES

1. W. Feld, *J. f. Gasbel.*, 1904, **47**, 545; H. E. Williams, "*Chemistry of Cyanogen Compounds*," 1915, 332.
2. H. T. S. Britton and E. N. Dodd, *J. Chem. Soc.*, 1932, 1945.

166, WICKHAM CHASE

WEST WICKHAM, KENT

March, 1939

The Routine Examination of Magnesium Trisilicate

By J. L. PINDER, B.Sc., F.I.C.

THE dispensing of substances for dealing with minor intestinal disturbances is the daily task of the pharmacist. Various alkaline powders, and materials such as chalk, charcoal and kaolin, are widely employed as antacids and as adsorbents* of toxins and alkaloids. Although a number of magnesium silicates have been used medicinally for a long time, it is only recently that they have been subject to critical examination and their uses defined. Mutch,¹ in an exhaustive survey, concluded that the most effective compound was an artificial silicate having the approximate composition $\text{Mg}_2\text{Si}_3\text{O}_8 \cdot n\text{H}_2\text{O}$. The silicate combines effectively the action of an antacid with that of an adsorbent for a wide range of materials, including dyes, alkaloids, colloids and toxins; in each category, however, a selective action is noticed, some dyes, for example, being much more effectively adsorbed than others at similar concentrations. With the probable increase in demand for trisilicate, tests for controlling its composition and adsorptive capacity become desirable. Analytical figures and a methylene blue adsorption test were put forward by Mutch in a second paper,² and Glass³ outlined further suggestions for analytical procedure, and gave a modified qualitative adsorption test.

The objects of the present investigation were: (a) to examine the chemical analytical procedure and to devise suitable routine tests; (b) to devise a modified quantitative adsorption test, capable, if possible, of correlation with Mutch's figures; (c) to determine whether or not the adsorption depends in any way on the physical condition of the trisilicate.

(a) ANALYTICAL PROCEDURE.—(1) *Magnesia and silica*.—The following methods were adopted after duplicate experiments on acid decomposition and fusion with alkali carbonate had shown fusion to have no advantage. The material (0.5 g.) was treated with hydrochloric acid, followed by evaporation, drying and boiling with dilute acid, and finally by filtration of the silica, which was ignited and weighed, as described by Treadwell and Hall.⁴ Re-evaporation, however, was omitted,

* The term "adsorbent" has been preferred to "absorbent" throughout this paper.

as without it the figures obtained are sufficiently accurate for routine purposes. The filtrate was made up to 250 ml. and the magnesium was determined in duplicate—volumetrically as the 8-hydroxyquinoline complex, and gravimetrically as pyrophosphate. For the oxine procedure, 25 ml. of the solution were treated as described by Mitchell and Ward,⁵ method (1), re-precipitation of the complex being unnecessary. To the boiling, faintly acid solution, was added 5 to 10 g. of ammonium acetate (AnalaR), followed by about 15 ml. of 2 per cent. oxine in 2 *N* acetic acid. The solution was then made faintly alkaline with ammonia (0.880) and the precipitate allowed to settle. The supernatant liquid was filtered off through a Jena G.3 glass filter, to which the precipitate was finally transferred and well washed with hot water. The precipitate was dissolved in 100 ml. of 2 *N* hydrochloric acid, 25 ml. of *N*/10 (*M*/60) potassium bromate and bromide solution were run in, and the excess was titrated back with *N*/10 thiosulphate solution after the addition of potassium iodide; 1 ml. of *N*/10 bromate \equiv 0.504 mg. of MgO. Results tend to be low in comparison with those obtained by the pyrophosphate method (usually 0.3 to 0.4 per cent. on the sample or 1 to 2 per cent. on the magnesium oxide), but this is offset by the greatly reduced time required, from $\frac{1}{2}$ to 1 hour. Two hundred ml. of the filtrate from the silica determination were used for the determination of magnesia as pyrophosphate.*

TABLE I

| Sample | SiO ₂ Per Cent. | MgO | | Ratio MgO: SiO ₂ | Moisture | | | Basicity ml. of <i>N</i> /10 HCl per g. |
|--------|-------------------------------|---------------------------------|--------------------|-----------------------------------|-------------------|-----------------------|--------------------|---|
| | | Pyro- phosphate Per Cent. | Oxine Per Cent. | | Free Per Cent. | Combined Per Cent. | Total Per Cent. | |
| 1a | 51.2 | 22.2 | — | 1:2.30 | 9.5 | 15.8 | 25.3 | 100 |
| 1b | 49.2 | 22.6 | 22.25 | 1:2.18 | 9.5 | 15.9 | 25.4 | 111 |
| 2 | 49.9 | 20.7 | 20.1 | 1:2.48 | 14.9 | 11.6 | 26.5 | 79.5 |
| 3 | 50.5 | 16.35 | 16.2 | 1:3.07 | 16.7 | 14.1 | 30.8 | 73.5 |
| 4 | 49.6 | 22.35 | 22.6 | 1:2.22 | 10.5 | 13.2 | 23.7 | 109.5 |
| 5 | 46.8 | 22.05 | 21.5 | 1:2.12 | 12.5 | 16.1 | 28.6 | 111 |

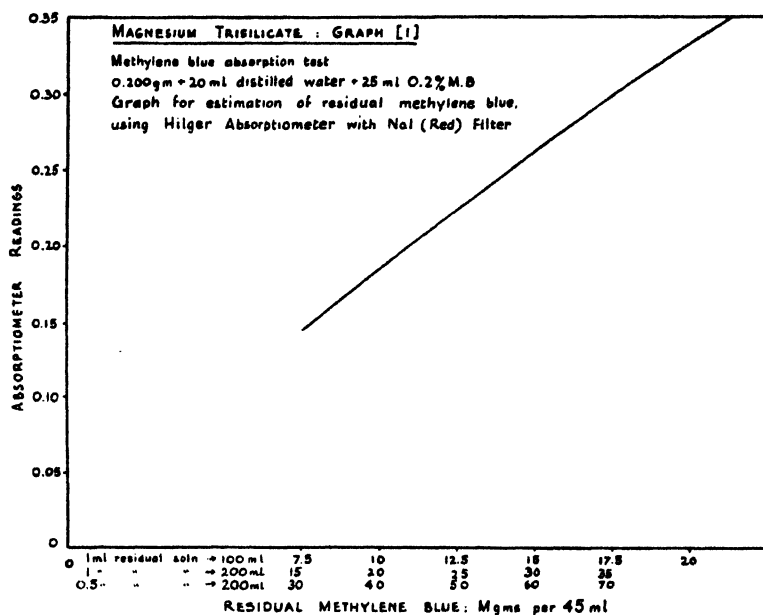
All results are calculated to sample "as received."

(2) *Moisture*.—The loss at 100° C. on 1 g. in about 4 hours was returned as free moisture, and the further loss on ignition as "combined" moisture.

(3) *Basicity*.—The procedure advised by Glass⁸ was adopted. Fifty ml. of *N*/10 hydrochloric acid were added to 0.2 g. contained in a 100-ml. stoppered flask, and the whole was shaken occasionally over a period of two hours. At the end of this time the excess acid was titrated back with *N*/10 caustic soda, with methyl orange as indicator. Results were expressed as ml. of *N*/10 acid per 1 g. of sample "as received." For analytical results, see Table I.

* A similar procedure is possible in the examination of tablets, granules, etc., containing magnesium trisilicate as a major ingredient. The finely-ground sample (0.5–1 g.) is gently ignited, if necessary, to remove organic matter, and then treated with acid as described. If the magnesium is estimated by the oxine procedure alone, sufficient filtrate remains from the determination of silica for the estimation of other inorganic constituents, e.g. precipitated chalk, which may be present.

(b) METHYLENE BLUE ADSORPTION.—(1) The method proposed by Glass³ cannot give quantitative results, owing to the difficulty of assessing the end-point. This is due to the fineness of some of the silicate particles, stained blue, which do not settle, thus giving a pale blue suspension, and also due to the adsorption of the dye by the glass of the flask, which is especially pronounced with poor samples of trisilicate because these have a low adsorptive capacity for the dye, and therefore leave a relatively large amount free to be adsorbed by the glass. Attempts to reduce suspension by centrifuging or filtration were unsuccessful. Similar experiments carried out in 100-ml. graduated flasks were slightly more successful. The flasks were shaken every half hour and the neck was observed immediately before each shaking. The same troubles were encountered, however, and the method was abandoned.



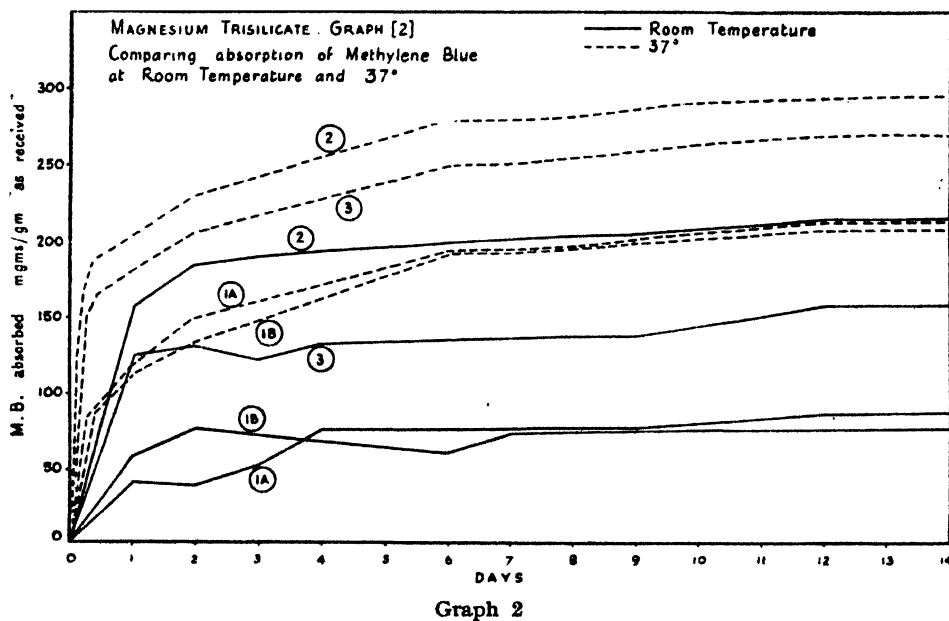
Graph 1

(2) A few experiments were made with a view to obtaining a figure that would represent the minimum amount of silicate required to adsorb a standard amount of dye in a standard time ($\frac{1}{2}$ hour). The relatively large quantities of silicate required obscured the end-point, owing to the suspension of silicate particles.

(3) Examination of Mutch's method. This method⁴ consists essentially in adding a standard amount of trisilicate to a standard amount of methylene blue in a given volume, and determining the residual dye after 14 days by a colorimetric method. For routine purposes a shorter test is desirable.

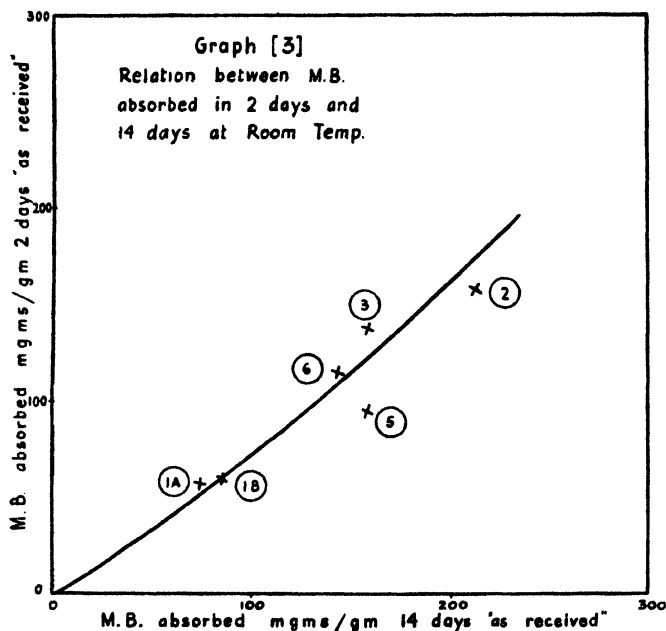
For this purpose investigations were made of (a) the relation between the adsorptions at room temperature and at a higher temperature—say, 37° C., and (b) the relation between the adsorptions in, say, 2 days and 14 days at room

temperature. The procedures used were as follows:—The sample (0.200 g.) was placed in a 50-ml. rubber-stoppered boiling-tube. For experiments at room temperature, 20 ml. of water followed by 25 ml. of a 0.2 per cent. aqueous methylene blue solution (B.D.H. \diamond SS) were introduced from a pipette, and the tube was closed and shaken periodically; 1 ml. or 0.5 ml. samples, depending on the observed degree of adsorption, were removed after shaking the tubes and immediately made up to volume, the unadsorbed methylene blue being measured on a Hilger Spekker Absorptiometer, using the No. 1 (red) filter. The experiments at 37° C. were made with 0.200 g. of material, 10 ml. of water and 35 ml. of methylene blue solution. The experiments at room temperature thus involved the use of 50 mg. of dye in 45 ml., and the experiments at 37° C., 70 mg. of dye in 45 ml. Graph 1 indicates suitable ranges of dilution for the determination of residual methylene blue. It is, however, necessary to calibrate each individual instrument separately.



(a) *Adsorption at Room Temperature and at 37° C.*—It was thought that, if a sample of trisilicate has a constant maximum adsorption of methylene blue, then this value might be more quickly reached at 37° C. than at room temperature. Four samples were examined, and it was found that the maximum adsorption figures (*i.e.* adsorption in 14 days) at the two temperatures were different. The time for the adsorption, at 37° C., of the amount of dye adsorbed in 14 days at room temperature was found also to be different with different samples; it was therefore concluded that the estimation of adsorption at 37° C. could not be correlated with the adsorption at room temperature (see Graph 2). During the course of this series of experiments, irregularities, especially with small degrees of

adsorption, *i.e.* high residual quantities of methylene blue, were observed in the graph. The cells were washed with distilled water before each determination, but it was observed that the methylene blue was adsorbed strongly on to the surface of the glass cell; ultimately it was found necessary to wash the cell with a few drops of conc. nitric acid, followed by distilled water, before each determination. Experiments were also made without the addition of methylene blue, after addition of an equivalent volume of water, to find the effect of the trisilicate suspension on the absorptiometer values. This was negligible and could be ignored.



Graph 3

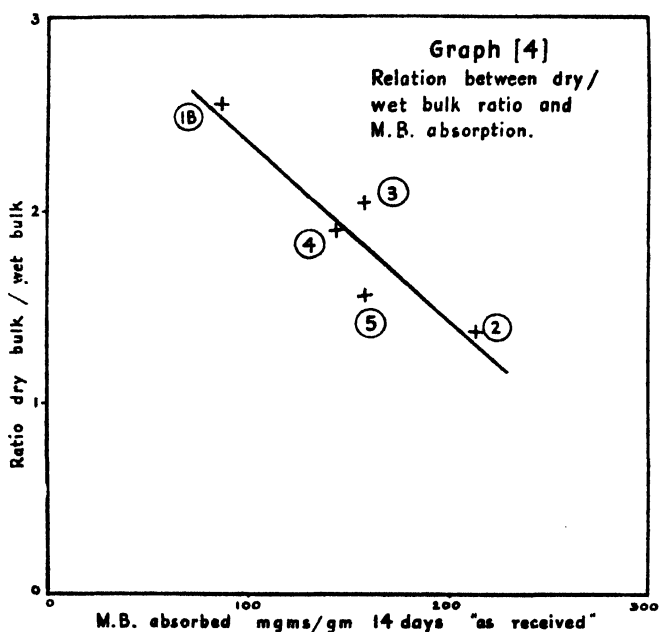
(b) *Relationship between Adsorption in 2 days and 14 days at Room Temperature.*

—The results obtained in 2 days and 14 days at room temperature were plotted (Graph 3); whilst no mathematical relationship is obvious, a general tendency is observable for the final adsorption figure to be about $1\frac{1}{2}$ times the adsorption at 2 days. From the calculated ratios, omitting sample (5) (see Table II), it would seem that samples showing a low adsorption in 2 days tend to show a higher ratio of adsorption in 14 days to adsorption in 2 days than samples showing a high adsorption in 2 days; in other words, "good" samples reach their maximum adsorption more quickly than inferior samples. Graph 3 is therefore probably non-linear, but this can ultimately be decided only after a considerably larger number of samples have been examined. If this is so, the 2-day adsorption test, besides taking less time than the 14-day test, permits of better differentiation between good and poor samples. Similar remarks apply to the test at room temperature compared with that at $37^{\circ}\text{C}.$: there is considerably better differentiation between good and poor samples at room temperature than at $37^{\circ}\text{C}.$ This is shown on Graph 2.

TABLE II

| Sample | Methylene blue adsorption, mg. per g. "as received" | | | | Ratio: room temp., 14/2 days | Ratio: 37° C., 14/2 days |
|--------|---|---------|--------|---------|------------------------------------|--------------------------------|
| | Room temperature | | 37° C. | | | |
| | 2 days | 14 days | 2 days | 14 days | | |
| 1a | 55 | 84 | 147 | 217 | 1.52 | 1.47 |
| 1b | 55 | 77.5 | 130 | 217 | 1.41 | 1.62 |
| 2 | 165 | 213 | 227 | 295 | 1.29 | 1.30 |
| 3 | 134 | 157 | 205 | 269 | 1.17 | 1.31 |
| 4 | 112 | 144 | — | — | 1.29 | — |
| 5 | 95 | 160 | — | — | 1.68 | — |

Some of the above figures are averages of duplicates.



Graph 4

(c) *Physical Properties.*—It was thought that adsorption might depend to some extent on physical condition, especially surface area per unit weight. The dry bulk of different samples was determined by weighing 10 g. on a slip of paper, breaking up lumps by smoothing with a watch glass, transferring to a 50-ml. graduated cylinder, shaking this vigorously for about 5 seconds, and then tapping it on a wooden bench about 25 times from a height of about $\frac{1}{4}$ inch; the volume was read after 1 minute. Water was introduced to bring the volume to the 50-ml. mark, with shaking, and the cylinder was allowed to stand for 2 hours: the wet volume was then read. In many instances no distinct line of demarcation was visible before two hours; after this period the apparent volume increased, possibly owing to hydration of the silicate. This is shown in Table III. No direct relationship between adsorption and either dry or wet volume is evident, but

samples with a high ratio of dry to wet volumes seem to have poor adsorption, and *vice versa*, as indicated by Graph 4.

TABLE III

| Sample | Dry bulk vol., ml. per 10 g. | Wet bulk, ml. occupied by 10 g. | | | | | Dry to wet ratio | 14-day adsorption mg. Meth. Blue per g. |
|--------|------------------------------------|---------------------------------|---------|---------|---------|---------|---------------------------|---|
| | | 2 hrs. | 5½ hrs. | 18 hrs. | 30 hrs. | 66 hrs. | | |
| 1b | 32 | 12.5 | 14 | 15.5 | 16 | 16 | 2.56 | 77.5 |
| 2 | 24.5 | 17.5 | 18.5 | 19.5 | 19.5 | 20 | 1.36 | 213 |
| 3 | 45.5 | 22 | — | 24.5 | 26.5 | — | 2.06 | 157 |
| 4 | 35.5 | 18.5 | 20.5 | 21.0 | 21.5 | 21.5 | 1.91 | 144 |
| 5 | 29.5 | 19 | — | — | — | — | 1.55 | 160 |

SUMMARY AND CONCLUSIONS.—(a) It is recommended that the analytical examination of magnesium trisilicate should include determinations of silica, magnesium, free and combined water and acid absorption. Methods are indicated for their determination.

(b) The merits of various times and temperatures for a methylene blue adsorption test are considered, and it is concluded that a two-day test at room temperature is of value. The figures thus obtained can be roughly correlated with those for adsorption after 14 days.

(c) The wet and dry bulks can be determined with advantage; the ratio of dry to wet bulk appears to have some relation with the results of the adsorption test.

I have to acknowledge the encouragement and help of Mr. A. F. Lerrigo, B.Sc., F.I.C., and to thank the directors of Glaxo Laboratories, Ltd., Greenford, for permission to publish this paper.

REFERENCES

1. N. Mutch, *Brit. Med. J.*, 1936, 1, 143, 205, 254.
2. ——— *id.*, 1937, 2, 735.
3. N. Glass, *Quart. J. Pharm.*, 1936, 9, 445.
4. Treadwell and Hall, "Analytical Chemistry," 8th Edition, Vol. II, pp. 427–8.
5. Mitchell and Ward, "Modern Quantitative Chemical Analysis," 1932, p. 103. Method (1).

ANALYTICAL DEPARTMENT

GLAXO LABORATORIES LTD.

GREENFORD, MIDDLESEX

October, 1939

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

DETERMINATION OF FAT BY THE GROSSFELD METHOD

GROSSFELD's method has been given particular prominence in recent German literature; special tables have been printed in connection with it, and arrangements have been made for the supply of the special apparatus for filtration without loss through evaporation. We recently tested the method, using the following simplified technique:—Five g. of maize meal were put into a 300-ml. Erlenmeyer flask, 50 ml. of trichloroethylene were accurately pipetted into it, and the flask was connected (rubber stopper) with a reflux condenser. The mixture was boiled for 7 minutes and then cooled, a little anhydrous sodium sulphate was added, and the contents of the flask were shaken and allowed to settle, after which 25 ml. were pipetted off through a cotton-wool guard plug, and filtered into a tared receiver, the paper being washed with a few ml. of the solvent. Two samples were worked on, and the results were checked by extractions with ether in a Soxhlet apparatus.

The dried fats from the Grossfeld process were appreciably darker than those from the Soxhlet extractions, and the Grossfeld results were very much higher.

A control test applied to the trichloroethylene (a "technical" product from a reputable firm) showed that it contained 0.065 g. of non-volatile matter per 100 ml. When allowance was made for this the results were as follows:

| | | Refined meal Per Cent. | Straight-run meal Per Cent. |
|--------------------|----|---------------------------|--------------------------------|
| Soxhlet extraction | .. | 1.2 | 4.2 |
| Grossfeld method | .. | 2.2 | 6.2 |

The discrepancies are very serious, and one wonders if rubber stoppers are as unaffected by trichloroethylene as has been assumed.

We should be grateful to learn the experiences of others who have used the method or who have employed Leithe's refractive technique.

W. H. KITTO

GOVERNMENT ANALYST'S LABORATORY
SALISBURY

SOUTHERN RHODESIA

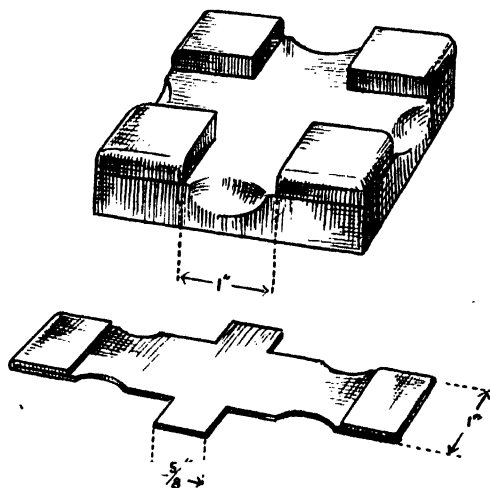
December 5th, 1939

TECHNIQUE FOR THE PREPARATION OF A STANDARD MICROSCOPE SLIDE, WITH PARTICULAR REFERENCE TO CHOCOLATE

IN the manufacture of chocolate it is well known that the size of the particles of sugar greatly influences the quality of the finished product. Measurement of the sugar particles has been mentioned by Knapp¹ in 1920, and further described by Bywaters² in 1930. Jensen³ described a microscope method he had applied successfully for a number of years, and a process is also given by Fincke.⁴ In these methods it is assumed that the microscope slide is made up in a standard way, but personal judgment is always required for the original amount of material taken. The subsequent manipulation has always been open to the appreciable

variations in technique introduced by different workers. With a view to preparing a slide in a standard way as free as possible from these variations I have devised the following method, which is applicable as a routine test.

PREPARATION OF STANDARD SLIDE.—To ensure that a standard amount of plain or milk chocolate is taken, a plate of 22-gauge metal drilled in the centre with a $\frac{5}{64}$ inch hole is used. The plate is placed on a microscope slide, and the hole is tightly packed with the material by means of the flat blade of a penknife. The excess of chocolate is removed by drawing the edge of the knife across the hole, any loose particles being removed by wiping with a finger or tapping the edge of the plate on the bench. The chocolate in the hole is poked out with a blunt steel tool which passes through the hole for $\frac{1}{8}$ to $\frac{3}{8}$ inch and is then stopped by a shoulder on the handle, which prevents the hole becoming enlarged in use. The chocolate should be deposited at the centre of the slide already used. The weight of chocolate given by this method is approximately 2.8 mg., but may vary by ± 0.2 mg., according to the nature of the material.



The definition of the sugar particles is increased by mounting the slide in red-coloured oil as suggested by Phillips.⁵ The oil is prepared by dissolving B.D.H. oil-soluble red in a small volume of trichloroethylene, filtering and adding the filtrate to arachis oil. The oil is added from a 5-mm. glass rod tapered to 3 mm. in a distance of 2 cm. and with the end rounded. In use, the first two or three drops are allowed to fall in the bottle and only a slow-forming drop which will fall clear of the rod is taken.

The slide is gently warmed, if necessary, excessive heating being avoided, and is then placed in the aluminium holder of the device shown in the diagram (Fig. 1), and the holder is moved until the oil and chocolate are in the centre of the "cross." A clean microscope slide is placed on top at right angles to the first slide and, without any additional pressure, the aluminium holder is moved six or seven times in each direction. The holder is designed so that the movement is $\frac{3}{8}$ inch.

The aluminium holder and the two slides are lifted out of the wooden part. The lower slide is held with the left hand and the upper with the right. The slides are parted, and the oily material clinging to the top slide is transferred to the bottom one by scraping the top slide against the edge of the bottom slide at an angle of approximately 45° . The major part can be removed on one edge and the

residual traces on the other. The material, now practically completely on the bottom slide, is collected to the centre of the slide by means of the edge of a sharp penknife, and a final mixing is given, only the edge of the knife still being used. Any material on the knife is then scraped on to the edge of the slide and as much as possible is transferred to the middle. A round cover-glass, $\frac{7}{8}$ inch in diameter, is placed on the top and allowed to sink by its own weight without external pressure of any sort.

The amount of material taken should be sufficient to spread to the edge of the cover-glass. A clear field with the particles sharply separated should be obtained, but, if not, it is permissible to move the cover-glass in two or three directions, but not exceeding $\frac{1}{16}$ in., by applying the point of a knife on the slide and not on the cover-glass. For ease of examination it is important that the cover-glass should be clean and free from smears, and industrial alcohol is recommended for the final cleaning.

If the cover-glass fails to sink down evenly, owing to the presence of a comparatively large particle of foreign matter, it is advisable to remake the slide from the beginning. It is important that no material be removed from the slide, and the details given must be strictly followed. Particular care should be taken in wiping slides and apparatus with a cloth, as the presence of fine hairs on the slide causes an uneven distribution of particles.

The amount of chocolate given by the plate described is suitable for finished chocolate, but for microscope examination during manufacture two other plates are used depending on the coarseness of the material. The three plates are as follows:

| No. | Thickness of plate Inch | Diameter of hole Inch |
|-----|----------------------------|--------------------------|
| 1 | 0.028 | 5/64 |
| 2 | 0.048 | 3/32 |
| 3 | 0.048 | 9/64 |

The plates are made of stainless steel or other metal resistant to wear and, for convenience, measure 2 in. by 1 in. The capacities of plates Nos. 2 and 3 are approximately $2\frac{1}{2}$ and 6 times that of plate No. 1 respectively. The plate to be used depends on the average size of the largest particles in the chocolate and is fixed as follows:

| Plate No. | Average size of largest particles |
|-----------|-----------------------------------|
| 1 | less than 0.0030 inch |
| 2 | 0.0030 to 0.0060 „ |
| 3 | above 0.0060 „ |

The amount of oil required for plates 2 and 3 is best found by experience, but is generally given by one free drop and afterwards touching the slide with the rod to give half a drop.

NOTES ON EXPERIMENTAL WORK.—The method described for preparing a slide takes $1\frac{1}{2}$ minute, and this compares favourably with the ordinary technique. To find if particles were broken down by the process, the lower slide was given 50 backward and forward movements instead of 6 or 7, but no difference in the particle size was detectable. Another variation comprised placing a 100-g. weight on the top slide in the holder, but even this had no breaking-down effect on particles, while after another test the two slides were inverted and the movement carried out in the opposite way. No difference was recorded. It was not found possible to dispense with the after-manipulation of the material, as the particles were insufficiently separated for measurement.

Tests were carried out to find what percentage of the material originally taken was secured under the cover-glass. It amounted to 75 per cent. with

plate No. 1 and 85 per cent. with plates 2 and 3. The figures were reasonably constant and, with care, different operators should retain approximately the same proportions.

CONCLUSION.—The main advantages of the method are that the same amount of chocolate is taken by different operators and that each slide receives the same amount of mixing. It is considered that the method should find application in other industries where particle-size measurement is carried out as a rapid routine test or where standard slides are required.

I wish to thank Mr. R. V. Wadsworth and Mr. J. R. Johnson for their interest and advice, and Messrs. Cadbury Bros. Ltd. for permission to publish this method.

H. C. LOCKWOOD

REFERENCES

1. A. W. Knapp, "*Cocoa and Chocolate*" (Chapman & Hall, 1920), p. 144.
2. W. H. Bywaters, "*Modern Methods of Cocoa and Chocolate Manufacture*" (J. & A. Churchill, 1930), p. 305.
3. H. R. Jensen, "*The Chemistry, Flavouring and Manufacture of Chocolate, Confectionery and Cocoa*" (J. & A. Churchill, 1931), p. 105.
4. H. Fincke, "*Handbuch der Kakaoerzeugnisse*" (Julius Springer, 1936), p. 456.
5. R. J. Phillips, *ANALYST*, 1921, **46**, 387.

CHEMISTS' DEPARTMENT

BOURNVILLE, BIRMINGHAM

November 5th, 1939

THE REACTION BETWEEN 2,2'-DICHLORODIETHYL SULPHIDE (MUSTARD GAS) AND BLEACHING POWDER

It has been observed that the reaction between mustard gas and bleaching powder is violently exothermic—so much so that the mixture frequently bursts into flame. It often happens, however, that a sample of the gas will inflame with one sample of bleaching powder but not with another. I recently tried a sample of specially good bleaching powder from a sealed tin freshly opened. This gave no sign of reaction at all with mustard gas, although several drops were added and even stirred in. On the addition of a few drops of water, however, the whole mass burst into flame. This seems to indicate that the failure of certain samples of bleaching powder to give a vigorous reaction is due to their having too low a moisture-content.

A. G. LIPSCOMB

68, BRIAN AVENUE

NORWICH

December 21st, 1939

AN ANCIENT EGYPTIAN MARKING INK

SEVERAL years ago I examined the marked characters on the winding sheet of Tchchuti Sat in the British Museum (B.M. 37,105), and gave an account of the pigment in *THE ANALYST* (1927, **52**, 27). This fabric was the one to which Budge refers in his book *The Mummy* (p. 137), where he says: "It is noteworthy that Egyptian ladies marked their linen with indelible ink." The pigment in this marking contained neither metals nor lampblack, but consisted of an organic colouring matter, possibly bistre, attached to the fibres of the fabric.

A still more ancient marking ink than that mentioned by Dr. Budge has recently been discovered, and I am indebted to Mr. A. Lucas for the opportunity of examining it. Three small pieces of linen fabric were found by Mr. W. B. Emery in the rubble filling of the shaft of a tomb of the Second Dynasty at Saggara, near Cairo (the ancient capital Memphis). The dynasty ended about B.C. 2980, and the tomb is probably of a much earlier date than that; it is not known who was buried in it. In Mr. Emery's opinion it is probable that the



Fig. 1

MARKING ON ANCIENT EGYPTIAN LINEN (x 1-5).

markings had some ritual significance, for, although the letters can be read, their meaning is not known. The fragments were cut, not torn, and were probably put intentionally into the shaft of the tomb.

As will be seen in the photograph (Fig. 1) of one of the three fragments, the linen fabric is very coarsely woven, and two of the sides end in a long rough fringe. The marked characters are of a brownish-yellow colour and are placed towards the edge of the fragment. The pigment dissolved with difficulty in hot hydrochloric acid and gave the usual reactions for iron. No other pigment could be detected, and it was concluded that the markings consist of iron oxide deposited on the fibres. The unintentional production of "iron mould" by laundries on modern linen suggests the way in which the iron oxide may have been fixed. The markings behave like iron oxide pigments in ultra-violet light, giving a black "fluorescence."

I have to thank Mr. T. J. Ward for his help in the microchemical examination of these markings.

C. AINSWORTH MITCHELL

12, LIVERPOOL TERRACE
WORTHING

January, 1940

Department of Scientific and Industrial Research

WATER POLLUTION RESEARCH

ANNUAL REPORT OF THE BOARD FOR THE YEAR ENDED JUNE, 1939*

THE Annual Report of the Water Pollution Research Board for the year ended June 30th, 1939, has been issued by the Department of Scientific and Industrial Research. In an introductory statement the Board points out that greater vigilance is required in protecting existing and future sources of water supply from undue contamination by the discharge of polluting substances; these measures are especially necessary during wartime, when there are movements of large numbers of people from one district to another. Research carried out under the supervision of the Board includes investigations on the treatment of water for domestic and industrial purposes, the treatment and disposal of sewage and trade effluents, and on problems of pollution of rivers.

BASE-EXCHANGE PROCESS OF WATER SOFTENING.—The base-exchange materials available for this purpose include natural glauconites, treated clays, synthetic zeolites, certain synthetic resins and materials prepared from carbonaceous substances such as coal. Of these materials, natural glauconites and treated clays have not hitherto been produced on a large scale in this country. Previous work under the Board has shown that satisfactory base-exchange substances can be obtained by treatment of fuller's earth, which occurs in large amounts in Great Britain. During the year under review, further work has been done on the examination of British minerals for the preparation of base-exchange materials. One of these minerals, a glauconitic sand, from a brick-works in Surrey, has a high base-exchange capacity and appears to be a suitable raw material for a base-exchange zeolite for the treatment of water. Other work on the base-exchange process has included the investigation of the effect of temperature on the base-exchange capacity of a number of representative base-exchange materials. Within the limits 4 to 20° C. temperature has no significant effect on the exchange capacity of any of the materials tested.

* Published by H.M. Stationery Office, York House, Kingsway, London, W.C.2. 1940. Price 1s. net.

LEAD IN DRINKING WATER.—One method of treatment of water to reduce its corrosive action on lead and other metals consists in passing the water through beds of pieces of limestone, marble or similar substances. This method is particularly attractive for rural districts and remote localities, since the plant, when properly designed, should require little skilled supervision. Several such plants are already in use in this country in rural areas, but much further information is required on the design and methods of operation of this type of plant, if satisfactory results are to be obtained, particularly in the treatment of certain types of water. The Board is making a systematic investigation of this problem. Suitable apparatus and methods of analysis were first developed for the determination of small quantities of lead taken up by water in passing through service pipes. Visits were then made to water-works where water is treated by passage through beds of limestone, and samples of the raw water and treated water were taken for analysis. Arrangements were then made to carry out experiments on the treatment of water in filters containing pieces of limestone, both in the laboratory and on a large scale. Many of the experiments are being made at the Blackmoorfoot Works of Huddersfield, where facilities have been provided by the Huddersfield County Borough Council.

BACTERIOLOGY OF FRESH WATER.—An investigation on the bacteriology of freshwater lakes and streams was begun in May, 1938; this work is being carried out for the Board by the Freshwater Biological Association at the Association's research station at Wray Castle, Ambleside, Westmorland. Preliminary work has indicated that fluctuations in the numbers of bacteria in the upper layers of water in Windermere can be correlated with the rainfall over the drainage area during the period immediately before the samples are taken. The increase in the numbers of bacteria in the lake after heavy rain, however, appears to be due to multiplication of bacteria already in the lake rather than to the washing in of soil bacteria by the storm water, since the types of bacteria isolated from the lake water are, in general, different from the types found in soil.

MILK FACTORY EFFLUENTS.—The Board's investigation of the problems of treatment and disposal of the highly polluting waste waters from milk-collecting and distributing depots, and from the manufacture of cheese, butter, and other products from milk, was begun five or six years ago. In 1935 two large experimental plants were erected at the milk depot and cheese factory at Ellesmere, Shropshire, where facilities were afforded by United Dairies Limited. Methods of treatment of the waste waters which had previously been investigated in the laboratory were tested in the two large-scale plants. These experiments, which were completed during the autumn of 1938, have proved that it is practicable to purify the waste waters either by the activated sludge process or by treatment in two percolating filters in series with periodic change in the order of the two filters. The conditions necessary for the satisfactory operation of these processes in the treatment of the waste waters from the various branches of the milk industry have been ascertained. Under normal factory conditions, the process employing two percolating filters in series is more economical and requires less supervision than the activated sludge process, and it produces final effluents of high quality. The milk industry has shown great interest in this investigation and has contributed a total of £12,050 towards the cost of the work.

TREATMENT AND DISPOSAL OF SEWAGE.—The method developed by the Board for the treatment of dairy waste waters in two percolating filters in series, with periodic change in the order of the filters, is in some respects an elaboration of the method used at many sewage disposal works, where settled sewage is passed through a single percolating filter. For a given volume of filtering material, however, the volume of dairy waste waters which can be treated daily by the new method is considerably greater than the volume of sewage which can be treated by the method of single filtration, allowance being made for the difference in

strength of the two liquids. Moreover, the quality of the effluent from the double-filtration process was much better than is usual at many sewage disposal works. This at once suggested the desirability of experiments on the treatment of domestic sewage by the double-filtration process. If the amount of sewage which can be efficiently treated by double filtration with a given volume of filtering medium is only 50 per cent. greater than by single filtration, the adoption of the new process should lead to considerable reductions in the extensions required at many sewage disposal works.

In addition to this work on the treatment of sewage by biological filtration, experiments have been continued in the laboratory on the treatment of sewage by the activated sludge process. Among the factors investigated were the effect of temperature, proportion of activated sludge, and the acidity or alkalinity of the mixture of sewage and sludge. Experiments have also been made on the effect of acids and alkalis, mineral salts, and organic substances, on the rate of sedimentation of fine particles from suspension in water.

Ministry of Health

ANNUAL REPORT OF THE CHIEF MEDICAL OFFICER*

IN the Twentieth Annual Report for the year 1938, Sir Arthur MacNalty reviews the nature of progress and the application of the conception to questions of health. This is followed by chapters on Vital Statistics, General Epidemiology Maternity and Child Welfare, The Relation of Food to Health and Disease, Milk, Control of the Purity of Food, Medical Intelligence and Research, Water Supplies, etc.

FOOD STANDARDS.—In the chapter on the Control of the Purity of Food the question of food standards is fully discussed. It is suggested that where standardisation is undertaken in connection with marketing schemes the object of which is to promote the sale of the higher grades and qualities, it may be of material assistance to fix standards for the higher rather than for the lower grades. "It is, however, questionable whether, from the public health viewpoint, standards should take account of the higher grades and qualities of food where the considerations are mainly aesthetic. It may be held that the primary object of food definitions and standards should be to exclude all articles the consumption of which is clearly prejudicial to the public interest, but not to furnish the vendor with a "selling point" for the higher and more expensive grades of food.

"In demands for standards emanating from commercial interests there is often discernible a desire to stifle competition. Frequently the grades or qualities to which objection is taken are sound wholesome articles of food, the suppression of which would be a distinct loss to the poorer classes of consumers. In such cases the consumer's interest may involve a standard sufficiently low to include nearly all the grades of the food in question provided that they are wholesome and nutritious, and provided also that a system of labelling is adopted which will ensure that the consumer knows what he is buying."

A statutory standard which lays down the composition of an article in numerical terms must be capable of being fully supported by analysis. It has been represented that in all cases where a standard is fixed it should be accompanied by a prescribed method, so as to ensure the results of different analysts agreeing. On the other hand, there are several objections to prescribing analytical technique, and it would seem best that, whenever possible, the analyst should be allowed a free hand to adopt the methods that he has personally found to be the best for the purpose, or which the general body of analysts may have agreed among themselves to adopt.

COPPER IN CONCENTRATED TOMATO PULP.—At a conference of Port Medical Officers of Health held in October, 1938, it was agreed that a tolerance of not more than 100 parts of copper per million in the dried total solids should be allowed until January 1st, 1940, and that a limit of 50 p.p.m. would have to be complied with on and after that date.

TOXICITY OF THE FLESH OF RABBITS KILLED BY HYDROGEN CYANIDE GAS.—At the request of the Ministry of Health laboratory experiments were made to ascertain if the use of hydrogen cyanide gas for the killing of rabbits would render the flesh unfit for human consumption. The rabbits were exposed for varying periods of time to high concentrations of the gas (up to 1 part in 200). The amounts of free cyanide in the blood and tissues varied with the time of exposure and the concentration. After exposure to a concentration of 1 in 2000 (which is quickly lethal) followed by exposure for 5 hours to the same concentration, small amounts of free cyanide were found in the edible portions of the rabbit. The highest amount found was 2 mg. in the body of a rabbit which had been exposed for 4 minutes to a concentration of 1 part in 200. It would appear that the flesh of rabbits exposed to concentrations of the gas probably higher than would ordinarily be employed cannot normally be regarded as unwholesome, but it would not be wise to say that in no circumstances would it be open to objection from the dietetic standpoint.

Rothamsted Experimental Station

REPORT FOR 1938*

THE Introduction to the Report contains a general account of the founding of the Experimental Station and of the development of its numerous activities. The Report itself gives a description of the work of the Station in the different laboratories and on the Woburn Experimental Farm. Among the interesting results discussed are the following:

USE OF TOWN REFUSE AS MANURE.—A prepared town refuse was compared with (1) sulphate of ammonia and (2) dung or rape dust, each given in single and double dressing, the nitrogen content being taken as the basis for comparison. The mean composition of the town refuse used was N, 0.82 per cent.; moisture, 30.3 per cent. The rates of application were: Single dressing of town refuse, about 5 tons per acre; rape cake, dung, 0.8 cwt. of N per acre. Single dressing of sulphate of ammonia, 0.4 cwt. of N per acre. Comparative tests with other nitrogenous manures were also made. The results (tabulated in detail) show that treated town refuse did almost as well as sulphate of ammonia providing half as much nitrogen, and was distinctly superior to sulphate of ammonia providing one quarter of the nitrogen, although it was much inferior to sulphate of ammonia supplying the same amount of nitrogen. The similarity in effectiveness to dung emphasises the desirability of further investigation (*cf.* E. Voelcker, *ANALYST*, 1939, 64, 510).

PHOSPHORUS COMPOUNDS IN SOIL.—Field experiments have indicated that, of the phosphate added as fertiliser, only about 25 per cent. is recovered in the crop in ordinary circumstances; the rest remains in the soil, but, in so far as can be discovered, in a form in which plants cannot easily take it up. Present evidence indicates that the soil is a poor storehouse for fertilisers.

MANGANESE DEFICIENCIES IN SOIL.—Three main types of soil are liable to manganese deficiency, as shown by characteristic crop troubles: (i) Neutral or alkaline soils, notably recently limed reclaimed heath soils, which do not contain

* Pp. 213. To be obtained from the Secretary, Rothamsted Experimental Station. Price 5s.

manganese mineral; these are liable to "grey speck" of oats. (ii) Alkaline fen soils; these are liable to "speckled yellows" of sugar beet. (iii) Heavily alkaline marsh soils, even if they contain manganese mineral; these are liable to "marsh spot" in seed peas. All these diseases have been remedied by suitable applications of manganese sulphate. It should be noted that they can all be brought on by over-liming.

COBALT DEFICIENCIES IN SOILS.—In the Chemical Department it has been shown that pastures of the Dartmoor area are deficient in cobalt; the sheep there suffer from a disease similar to that in New Zealand (*cf.* ANALYST, 1938, 63, 112). The remedy is to give a cobalt lick, but it is clearly desirable to make a survey of other hill or moorland grazings.

X-RAY ANALYSIS OF SOIL MINERALS.—X-ray analysis is now used in the Chemical Department for the identification of the minerals in the different soil fractions, and special attention has been devoted to the clay fraction. The special properties of this fraction are largely due to certain components now under investigation. They are very complex, and their smallest particles are shown by X-ray analysis to consist of a lattice structure in which layers of silicon and oxygen atoms alternate sandwich-like with layers of aluminium and oxygen atoms arranged systematically. The particles are electrically charged and hence have associated with them various ions, of which the most important from the point of view of soil fertility are calcium, and, in the Rothamsted conditions, hydrogen and potassium, but in arid conditions sodium and magnesium. These cations are replaceable by others: the "souring" of soil is caused by the replacement of calcium by hydrogen; conversely the "sweetening" of soil by liming is due to the replacement of hydrogen by calcium. An account is given in the Report of the three ways in which the electric charges appear to originate.

COLOUR OF THE SOIL.—Soil surveyors use the colour of the soil as one of its properties for classification, but the estimation of soil colours is very vague. Dr. Schofield has devised an instrument for the exact measurement of colour, and this has been taken over by Tintometer Limited.

WATER SUPPLY TO PLANTS.—A method of measuring the intensity with which soils hold water has been worked out in the Physics Department. The underlying conception of water suction is being applied to a study of the pore size distribution in soils.

CROPS AND MICRO-ORGANISMS.—*Clover Nodule Bacteria.*—Some strains of clover nodule bacteria are much less efficient than others. One of the poorest, found on the Welsh hills, has been studied in detail. It is so inefficient that it can barely sustain its host plant. The reason for this has been traced to some incompatibility between them and their host. Evidence has been obtained that plants bearing inefficient nodules produce some substance toxic to the plant, so that while they begin by fixing nitrogen just as the more efficient forms do, in a very short time the nodules begin to disintegrate.

The Denitrification Process.—The process of denitrification whereby nitrates are reduced in the soil to gaseous nitrogen has hitherto been regarded as entirely anaerobic. It is now shown that this is not so, and that complete reduction of nitrate to gaseous nitrogen can take place under anaerobic conditions, with the difference that, for a C/N ratio of 10, the whole of the carbon supplied is used up under aerobic conditions, but part of it is left untouched under aerobic conditions.

THE CABBAGE APHIS.—It has been found that the rate of reproduction of cabbage aphides is dependent on the composition of the cabbage, and that the aphides themselves affect not only the yield but also the composition of the cabbage.

VIRUS DISEASES.—From plants infected with two strains of potato virus "X," nucleo-proteins have been isolated which in many respects resemble those previously obtained from plants infected with tobacco mosaic virus. When

precipitated from solutions with acids or salts these proteins are amorphous. From plants infected with tomato bushy stunt virus another nucleo-protein has been isolated; after precipitation with salts this crystallises in rhombic decahedra. It is the first virus to be isolated in a fully crystalline state. It differs from those previously studied in having spherical instead of rod-shaped particles; also in having a much greater nucleic acid content.

Other sections of the Report deal with Insecticides and Fungicides, Fungus Diseases of Crops, Statistical Control of Experiments, Field Experiments at Outside Centres, etc.

The Sale of Milk Regulations, 1939

STATUTORY RULES AND ORDERS, 1939, No. 1417*

ADULTERATION. FOOD AND DRUGS ACT, 1938

THE Minister of Agriculture and Fisheries, in exercise of the powers conferred on him by Section 23 of the Food and Drugs Act, 1938, does hereby make the following Regulations:

Milk

1. Where a sample of milk (not being milk sold as separated, or condensed, milk) contains less than 3 per cent. of milk-fat, it shall be presumed for the purposes of the Food and Drugs Act, 1938, until the contrary is proved, that the milk is not genuine, by reason of the abstraction therefrom of milk-fat, or the addition thereto of water.

2. Where a sample of milk (not being milk sold as separated, or condensed, milk) contains less than 8.5 per cent. of milk-solids other than milk-fat, it shall be presumed for the purposes of the Food and Drugs Act, 1938, until the contrary is proved, that the milk is not genuine, by reason of the abstraction therefrom of milk-solids other than milk-fat, or the addition thereto of water.

Separated Milk

3. Where a sample of separated milk (not being condensed milk) contains less than 8.7 per cent. of milk-solids other than milk-fat, it shall be presumed for the purposes of the Food and Drugs Act, 1938, until the contrary is proved, that the milk is not genuine, by reason of the abstraction therefrom of milk-solids other than milk-fat, or the addition thereto of water.

Commencement

4. These Regulations shall come into operation on the first day of October, one thousand nine hundred and thirty-nine.

Extent

5. These Regulations shall extend to England and Wales.

Short Title

6. These Regulations may be cited as the Sale of Milk Regulations, 1939.

In witness whereof the Official Seal of the Minister of Agriculture and Fisheries is hereunto affixed this 1st day of October, one thousand nine hundred and thirty-nine.

(Signed) DONALD FERGUSON
(Secretary).

* H.M. Stationery Office, York House, Kingsway, London, W.C.2. 1939. Price 1d. net.

British Pharmacopoeia Commission

REPORT OF THE COMMITTEE ON PHARMACY AND PHARMACOGNOSY*

SECTION I.

The Sub-Committee on Crude Drugs have reviewed the 82 monographs submitted to them by the Commission, and have rewritten them so as to present a more complete account of the microscopy of commercial drugs than has been given in previous Pharmacopoeias, and also to indicate in sufficient detail the diagnostic microscopical features of the powders. Four complete monographs (*Belladonnae Folium*, *Belladonnae Radix*, *Cinchona*, and *Nux Vomica*) are included in the Report in order to show the general scope and mode of presentation. As regards the other monographs it is thought that a concise statement of the standards and tests for purity should be sufficient for the present purpose.

TYPES OF STOMATA.—The Sub-Committee recommend that, for reference in relation to the microscopy of drugs, the following definitions (applicable only to mature stomata) should be included in an Appendix to the Pharmacopoeia.

Ranunculaceous Type—with no special subsidiary cells.

Cruciferous Type—often with three or more accessory cells, one of which is distinctly smaller than any of the others.

Rubiaceous Type—often with two subsidiary cells, with their long axes parallel to the pore.

Caryophyllaceous Type—often with two subsidiary cells, lying round the ends of the guard-cells.

STANDARDS AND TESTS FOR PURITY.—*Drying of Drugs.*—The Sub-Committee recommend that all determinations for ash, acid-insoluble ash and water-insoluble ash should be made on the drug as received (air-dried drug).

Volatile Oil.—A method has been published in Report No. 11.

Powders.—The revised monographs are arranged to give, in most instances, descriptions for the powders of crude drugs. For the present the assumption is made that it will be possible to apply the standards for foreign organic matter to the official drugs in the form of powder. It is thought advisable for the present to restrict the standards for volatile oil in crude drugs to the drugs in the unground condition, with the exception of powdered cinnamon, for which a minimum content of oil is proposed (not less than 0.7 per cent.). Investigations are in progress to ascertain the loss of volatile oil during powdering and storage of powders.

Foreign Organic Matter.—This term should be used, in preference to "other organic matter" to designate organic matter which does not form part of the drug as defined by the monograph.

SUMMARY OF THE REQUIREMENTS FOR INDIVIDUAL DRUGS.—Standards and tests for identity are given for 78 monographs of the current Pharmacopoeia.

SECTION II.

The Sub-Committee on Extracts, Liquid Extracts and Tinctures have reviewed the monographs in the current Pharmacopoeia and recommend that the general principles there followed should be continued, *vis.* that instructions should be given for the preparation of relatively small quantities of these galenicals, and that recognition of deviations from the strict details of the pharmacopoeial monographs should be given in General Notices to the Pharmacopoeia.

Certain changes are recommended in the following monographs:—*Extractum Belladonnae Liquidum*, *Extractum Ipecacuanhae Liquidum*, *Extractum Sennae Liquidum*, *Tinctura Capsici*, *Tincturae Nucis Vomicae*.

New monographs are proposed for *Extractum Hamamelidis Siccum* and *Tinctura Gelsemii*.

SECTION III.

The Sub-Committee on Waters, Infusions, Solutions, Spirits and Syrups recommend certain changes in 14 monographs. New monographs are proposed for *Liquor Sodii Chloridi Compositus* (Ringer's Solution), *Spiritus Lavandulae*, and *Syrupus Codeinae Phosphatis*.

SECTION IV.

The Sub-Committee on Ointments and Miscellaneous Galenicals recommends certain changes in 14 monographs. New monographs are proposed for *Acriflavine Preparations*, *Emulsio Olei Morrhuæ*, *Emulsio Paraffini Liquidum*, *Glycerinum Acriflavinae*, *Injectio Calcii Gluconatis*,

* Report No. 13. Published by authority of the General Medical Council, 44, Hallam Street, London, W.1. September, 1939. Price 2s. 6d.

Injectio Procainae et Adrenalinæ, Injectio Quininae et Urethani, Injectio Sodii Morrhuae, Linctus Diamorphinae, Linimentum Aconiti Oleosum, Linimentum Methylis Salicylatis, Lotio Acriflavinae, Lotio Calaminae, Pasta Acidi Tannici, Pillulae Digitalis Compositae, Pulvis Bismuthi Compositus, Pulvis Catechu Compositus, Pulvis Kino cum Opio, Suppositoria, Suppositorium Adrenalinæ, Unguentum Gallae cum Opio, Unguentum Hamamelidis, Unguentum Hydrargyri Dilutum, Unguentum Methylis Salicylatis Compositum.

Mercury Ointments.—The difficulties that have arisen from the use of various names and synonyms for ointments containing metallic mercury have been considered, and the Sub-Committee recommend:

- (a) That the monograph on the Ointment of Mercury of the current Pharmacopoeia, containing 30 per cent. of mercury, should be continued without the synonym "Mercury Ointment."
- (b) That a Diluted Ointment of Mercury, containing 10 per cent. of mercury, should be introduced with the synonym "Blue Mercury" and with the addition of a note explaining the application of the names "Mercury Ointment" and "Mercurial Ointment." The draft for "Blue Ointment" submitted contains one-third of Mercury Ointment and two-thirds of Benzoinated Lard.

When Mercury Ointment or Mercurial Ointment is prescribed or demanded, Diluted Ointment of Mercury shall be dispensed or supplied, unless, on enquiry, it is ascertained that Ointment of Mercury is required.

SECTION V.

The Sub-Committee on Tablets has explored the possibility of defining tablets and the general principles have been worked out, but further investigation is required before definite conclusions can be reached.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Freezing-point of Milk. L. M. Lampert. (*J. Assoc. Off. Agr. Chem.*, 1939, **22**, 768-771.)—Hortvet (*J. Ind. Eng. Chem.*, 1921, **13**, 198) reported that the depression of the freezing-point of pure milk, as determined by the instrument he described, now widely used, ranged from 0.534° to 0.562° C., with an average value of 0.548° C. It would appear that the currently accepted average figure was obtained by rounding off this figure to 0.55° C. Bailey (*J. Assoc. Off. Agr. Chem.*, 1922, **5**, 484), reporting collaborative work on some 2700 samples, gave an average value of 0.544° C. Results from the Union of South Africa (*ANALYST*, 1937, **62**, 44) gave a range of 0.528° to 0.561° C. and an average of 0.541° C. Stubbs and Elsdon (*ANALYST*, 1934, **59**, 146) found for 1000 samples a range of 0.529° to 0.563° C. and an average of 0.544° C. The freezing-point depressions of 24 carefully authenticated samples from large and small herds in different parts of California were determined with minute attention to the details of the process and to the calibration and standardisation of the thermometers, and the average value found was 0.536° C. A survey of all the data obtained and other recently published data indicated that the accepted average depression of 0.550° C. is somewhat high. The authorities of New South Wales (*ANALYST*, 1937, **62**, 610) have considered it advisable to accept 0.535° C. and those of Western Australia (*ANALYST*, 1938, **63**, 890) to accept 0.540° C. as the average depression of pure milk. The data obtained by Hortvet, Bailey, and Stubbs and Elsdon were combined and examined statistically. The distribution curve was found to be normal and very

symmetrical, with a range of 0.523 to 0.566° C., a mean of 0.544° C., and a median of 0.543° C. Out of 1224 values, 54 per cent. showed a depression of 0.544° C. or less and 84.7 per cent. a depression of 0.550° C. or less. Since the depression of the freezing-point of milk from healthy cows has been proved to be unaffected by the season of the year or the feed, these values may be assumed to be characteristic of much of the milk produced. It is, therefore, suggested that, when control samples are not available, the value 0.540° C. be accepted as the average freezing-point depression of pure milk, especially when the results are to be used for the detection and determination of added water.

A. O. J.

Rapid Method for the Determination of Chlorides in Tomato Products.

L. M. Beacham. (*J. Assoc. Off. Agr. Chem.*, 1939, **22**, 765-766.)—The following method avoids such difficulties as slow evaporation in the official method for the determination of chlorides in tomato products ("*Methods of Analysis*," A.O.A.C., 1935, 500) and slow filtration in the tentative method for their determination in tomato juice (*J. Assoc. Off. Agr. Chem.*, 1937, **20**, 78) and gives excellent results. The tomato product (5 g.) is shaken with about 50 ml. of 80 per cent. alcohol, and after the addition of 1 ml. of conc. nitric acid and 25 ml. of *N*/10 silver nitrate solution the mixture is diluted to 100 ml. with the alcohol, thoroughly mixed, and centrifuged at 1800 r.p.m. for 5 minutes. A 50-ml. portion of the supernatant liquid is titrated with *N*/10 ammonium thiocyanate solution in presence of 2 ml. of saturated ferric ammonium sulphate solution in the usual manner. Results obtained by this method agreed well with those obtained by the official method. Tomato pastes and juices were prepared and their natural chloride-content was determined by this method. Known amounts of salt were then added, and the determination was repeated. The results obtained agreed closely with the known amounts of salt present.

A. O. J.

Estimation of Soya-bean Flour in Sausage by determining Non-fermentable Sugars. **W. B. Hendrey.** (*Ind. Eng. Chem., Anal. Ed.*, 1939, **11**, 611-613.)—A direct carbohydrate method based on hydrolysis to reducing sugars and the determination of these by Fehling's solution is put forward for the determination of soya-bean flour in sausage. The sample should contain about 2 g. of soya-bean flour (50 g. of sausage). It is prepared as directed by the A.O.A.C. (grinding with alcohol, extraction with petroleum spirit and re-grinding), and the water-soluble sugars are removed by washing the weighed sample by decantation with neutralised 50 per cent. alcohol. The liquid is poured through an 11-cm. 42 Whatman paper coated with 0.3 cm. of washed Filter-cel, and most of the solid is transferred to the paper and washed five times with the alcohol. If desired, the determination of soluble added sugars may be made. The insoluble sugars are then determined. After partial drying the washed residue and Filter-cel are transferred to the original flask, and the paper and sides of the flask washed with 120 ml. of 2.5 per cent. hydrochloric acid. The flask is fitted with rubber-stoppered reflux air condensers and held in a boiling saturated salt-bath for 3 hours. The liquid is nearly neutralised with 10 per cent. sodium hydroxide solution and transferred to a 250-ml. flask, hot water is added nearly to the mark, followed by 2 ml. of 50 per cent. phosphotungstic acid, and the liquid is made up

to 250 ml. with water, shaken and centrifuged, and tested for complete precipitation. Two hundred ml. are then pipetted into a 250-ml. flask and potassium chloride is added in slight excess to precipitate the excess of phosphotungstic acid, followed by 1 drop of methyl orange solution, and the liquid is neutralised to methyl orange and litmus, cooled, made up to volume, shaken and filtered. If desired, the percentage of insoluble sugars may be determined in an aliquot portion by the A.O.A.C. method, and for soya-bean will be about 11 to 12 per cent. of invert sugar or 10.5 to 11.5 of dextrose. The non-fermentable sugars are determined by a quick fermentation method adapted from Bailey (*Conn. Agric. Expt. Sta. Bull.*, 1937, 401, 869). Bakers' yeast is washed 5 times by stirring with 3 times its volume of water and centrifuging, and a 25 per cent. suspension in water is kept at about 0° C. Ten ml. are placed in a 100-ml. centrifuge tube and centrifuged, the water is poured off, and the inside of the tube is dried with filter paper. About 60 ml. of the sugar solution are added (diluted if necessary so that not more than a 0.1 per cent. concentration of fermentable sugars is present), stirred and left for 45 minutes, preferably at 30° C., with one stirring during this period. The solution is centrifuged and filtered, and the sugar is determined as invert sugar by the A.O.A.C. method; the percentage of non-fermentable sugars, multiplied by 9.4, gives the percentage of soya-bean in the sample. If lactose is absent the method may be shortened, but the short method suggested for the determination of total non-fermentable sugars is at present only tentative, since sufficient data are not yet available for determining the general validity of the blank correction.

D. G. H.

Indirect Volumetric Determination of Alkaloids. G. Auguste. (*J. Pharm. Belg.*, 1939, 21, 935-941, 961-964.)—Several attempts have been made to employ modifications of Mayer's reaction with alkaloids as quantitative methods, the most successful being that of Jonesco-Matiu and Varcovici (*J. Pharm. Chim.*, 1926, 8, 533; Abst., ANALYST, 1927, 52, 100) by which the alkaloidal iodo-mercurate is separated and decomposed by means of nitrous and sulphuric acids and its mercury-content determined by the procedure of Votoček and Kaspárek (*Bull. Soc. Chim.*, 1923, 33, 110; Abst., ANALYST, 1923, 48, 192). Since the alkaloids tend to form more than one iodo-mercuric salt (François and Blanc, *Bull. Soc. Chim.*, 1922, 31, 1208, 1304; cf. Abst., ANALYST, 1922, 47, 440), such methods have obvious disadvantages. Potentiometric methods for the titration of the iodo-mercuric anion (e.g. that of Maricq, *Bull. Soc. Chim. Belg.*, 1929, 38, 259; 1930, 39, 496; Abst., ANALYST, 1930, 55, 284; 1931, 56, 120) have not found general acceptance. In the reaction between alkaloids and potassium mercuric iodide in an acid medium, formation of the alkaloidal hydriodide that combines with mercuric iodide must entail a reduction in the acidity of the system equivalent to the amount of alkaloid present. This principle has now been applied to the determination of alkaloids by an acidimetric method. The reagent was prepared by diluting a solution of 25 g. of potassium iodide and 36 g. of mercuric iodide in 200 ml. of air-free water to 1 litre and allowing the solution to stand overnight before filtering. Fifty to 100 mg. of atropine were dissolved in 10 to 20 ml. of *N*/10 sulphuric acid, and the reagent was added, drop by drop, from a burette,

coagulation of the yellow colloidal precipitate being induced by agitation after each addition. When precipitation was complete the amount of reagent added was noted, and an excess of four to eight times this amount was run in. The mixture was vigorously shaken, allowed to stand until the supernatant liquid became clear, and, after addition of a little barium sulphate, was filtered through a double or triple filter-paper. An aliquot portion of the filtrate was titrated with *N*/10 sodium hydroxide solution, phenolphthalein being used as indicator. The reagent (20 ml.) was then mixed with 10 ml. of *N*/10 sulphuric acid and titrated with *N*/10 sodium hydroxide solution in the same manner. The amount of atropine present was calculated from the reduction of acidity caused by the formation of the complex salt. Each ml. of *N*/10 acid is equivalent to 0.0289 g. of atropine. The burettes used in the final titrations were graduated in 1/50th and 1/100th ml. The relative error of the method was usually not more than 1 to 2 per cent.; in rare instances it reached 3 per cent. The results of the acidimetric and iodimetric methods agreed well, the relative errors being of the same order as that of the iodomercurate method. Further investigation is being made to apply the method to other alkaloids and to the determination of alkaloids in galenical preparations.

A. O. J.

Application of the Herapathite Reaction to Aristoquin. M. Wagenaar. (*Pharm. Weekblad*, 1939, 76, 1544-1545.)—Methods for the detection of aristoquin (diquinine carbonic ester), $\text{CO}(\text{OC}_{20}\text{H}_{23}\text{N}_2\text{O})_2$, are based on the assumption that when this substance is dissolved in hydrochloric or sulphuric acid, carbon dioxide is evolved, and quinine chloride or disulphate is formed, respectively (*cf. id.*, 1934, 71, 316). The author provides evidence that in practice this is not entirely correct. Thus quinine sulphate has an intense blue fluorescence, whilst a solution of aristoquin in sulphuric acid fluoresces with a greenish-blue colour. The difference is apparent without the aid of an ultra-violet lamp, and is conveniently demonstrated by focussing sunlight, with the aid of a lens, on to the test-tube containing the liquid. Similarly, the herapathite reaction (whether used to produce the complex sulphate or selenate) is difficult to demonstrate for a solution of aristoquin in acid, whereas it is very sharp for pure quinine salts. The author has shown (*id.*, 1929, 66, 177) that the presence of acetone usually induces crystallisation in the herapathite reaction. This, however, does not apply to solutions of aristoquin in acid, unless the quinine salt so formed is first purified (*e.g.* by precipitation); the greater the degree of purity, the sharper the herapathite reaction. It is concluded, in fact, that the above dissimilarities between quinine salts and solutions of aristoquin in acids are due to the presence of impurities in the latter, and that these result from the method of preparation of aristoquin, namely, by the reaction of diphenyl carbonate and quinine; they may be due to the presence of an excess of the former or of the products of side-reactions. Another method of inducing the herapathite reaction is as follows:—The sample is evaporated slowly with a dilute solution of sodium hydroxide, and 1 drop of a solution of iodine in potassium iodide is added to a solution of the residue in dilute sulphuric acid. The brown precipitate which results is dissolved in acetone, and when the yellow solution so produced is evaporated the crystals of herapathite form rapidly. This method

serves satisfactorily as a test for aristoquin, and, although the crystals are not so well-formed as those produced from a pure quinine salt, they enable a strong dichroism to be detected.

J. G.

Reactions for the Identification of Isazin. M. J. Schulte. (*Pharm. Weekblad*, 1939, **76**, 1256–1257.)—Isazin is diacetyldihydroxy-phenylisatin, and the reactions described indicate the presence of the phenol, isatin and acetyl groupings. (1) The phenolic group is shown by Ehrlich's diazo-reaction. A solution of 10 mg. of the sample in 1 ml. of conc. alcohol is made just alkaline with 1 drop of sodium hydroxide solution and, on the addition of 1 drop of a 0.5 per cent. solution of sodium nitrite, and sulphanilic acid, an orange-red colour develops if isazin is present. On acidification with sulphuric acid the colour becomes yellow, and, on warming, an odour of ethyl acetate is evolved. (2) To a solution of 10 mg. of sample in 1 ml. of conc. alcohol is added 1 ml. of 0.1 *N* sodium hydroxide solution. On warming, a pale violet colour is obtained, and, on subsequent cooling and addition of 1 drop of 0.5 *N* bromine water, a dark blue colour results. If the solution is then shaken with chloroform the blue colour appears in the chloroform layer, the water layer being violet. Evidence is provided in support of the view that the blue compound is a bromine substitution-product of isatin. The reaction is successful only in weakly alkaline solutions and, consequently, the bromine water must not be too old. In presence of acid the blue colour changes to yellow, and if an excess of bromine water is present the colour is dirty green, and a yellow-brown precipitate is deposited. The blue compound is an indicator, which turns violet in alkali and yellow in acid, and it may be obtained by evaporation of the chloroform. A solution of the residue in alcohol gives a sharp end-point in the titration of borax with 0.1 *N* acid. (3) If 1 drop of a 0.1 per cent. solution of potassium permanganate is added to the violet alkaline alcoholic solution (see above) a dark purple-red colour is developed. This is preferable to the reaction with bromine water, in that it is less affected by outside influences. (4) A solution of 10 mg. of isazin in 1 ml. of conc. sulphuric acid develops a purple colour.

J. G.

Biochemical

Inactivation of the Enzymes of Gum Arabic. J. P. Kieft. (*Pharm. Weekblad*, 1939, **76**, 1133–1136.)—The enzymes of gum arabic are usually inactivated by heating a solution of the gum to 100° C. or by preparing *Gummi arabicum resiccatum* by evaporation of the mucilage. Thus, according to Laursen (*Dansk Tidsskrift for Farmaci*, 1932, p. 54) the oxidases and peroxidases are inactivated after 15 and 30 minutes, respectively, at 80° C. Since it has been accepted that, in general, enzymes are more stable to heat-treatment when in the dry state than in presence of moisture, the following experiments were carried out with the object of demonstrating the effect, on the inactivation, of variations in the technique of the method used. In each instance 500 mg. of the powdered gum (or an equivalent quantity of the mucilage) were dissolved in 3 ml. of water, and the solution was shaken with 4 drops of a 5 per cent. tincture of guaiacum; the

time that elapsed before a blue colour was produced was then noted, and these values are given below:—(1) Original gum arabic, 4 minutes. (2) Ten g. of gum were heated in a closed 15-ml. flask in a current of steam at 100° C. for 30 minutes, the flask being at 94° C. for 15 minutes; 70 minutes. (3) Experiment (2) was repeated, except that a boiling water-bath was used in place of the steam; 70 minutes. (4) The gum (50 g.) was heated in a 75-ml. flask on a water-bath, so that the temperature rose to 92° C. in 20 minutes, and was maintained at 92° C. for 2 hours; 105 minutes. (5) The powdered gum was heated in a flat dish in a drying-oven at 103° to 105° C., and the oxidase-values were determined after 0.5, 1, 2, 3.5 and 5 hours; 7, 20, 20, 25 and 25 minutes, respectively. (6) Gum arabic mucilage was heated for 1 hour at 70° and 100° C.; 35 minutes, and no change after 120 minutes, respectively. (7) *Pulvis Gummi arabici resiccat.* was prepared by evaporating the mucilage in air in a flat dish on a boiling water-bath at 65° to 80° C., this being completed in 1 hour; 70 minutes. (8) A portion of the product obtained in (7) was heated for 10 minutes at 103° to 105° C. in a drying-oven; no change after 120 minutes. (9) *Pulvis Gummi arabici resiccat.* was prepared by distillation in a vacuum, on a water-bath, in a stream of carbon dioxide for 1 hour at 75° to 85° C.; 30 minutes.

J. G.

Phytic Acid and the Rickets-producing Action of Cereals. D. C. Harrison and E. Mellanby. (*Biochem. J.*, 1939, 33, 1660–1680).—It has long been known that certain cereals, especially oatmeal, promote rickets in young animals. This rachitogenic activity is not the result of a deficiency of calcium or phosphorus in the cereal, as oatmeal for instance contains large amounts of both these elements. At first the presence of an “anti-calcifying toxamin” was postulated, but later, Bruce and Callow suggested that the rickets resulted from the non-availability of the phosphorus, which in such cereals, is present chiefly in the form of phytin (calcium magnesium inositol hexaphosphate). These workers, however, used diets rich in calcium and poor in phosphorus, whereas it has now been found that oatmeal is equally rachitogenic when fed to puppies on a diet having a normal calcium/phosphorus ratio. Under these conditions an adequate amount of phosphorus is present, so that the above explanation no longer holds good. Similar rachitogenic activity is also possessed by sodium phytate and phytic acid, but not by commercial phytin (the calcium magnesium salt), which may even be slightly antirachitic. The rachitogenic factor was isolated as follows:—Defatted oatmeal was hydrolysed by diastase to destroy the starch and then extracted with cold hydrochloric acid. Phytic acid was isolated from this extract by precipitation as the insoluble iron salt, and this was converted into the sodium salt. The latter was found to have the same rachitogenic activity as the oatmeal from which it was prepared; moreover, the activity was not lost on purification. The rachitogenic activity of oatmeal and of phytic acid and sodium phytate was inhibited by adding sufficient calcium. The hypothesis is now advanced that the rickets is produced by the phytic acid immobilising the calcium in the oatmeal and possibly also in other food eaten at the same time, by inhibiting its absorption from the alimentary canal.

F. A. R.

Colour Reaction of Methionine. J. J. Kolb and G. Toennies. (*J. Biol. Chem.*, 1939, 131, 401-407.)—When a solution of cupric chloride in conc. hydrochloric acid is added to methionine a colour is produced which resembles that of iodine-iodide solutions, varying from a deep brown to a pale yellow according to the concentration of methionine. This reaction was not obtained with twenty other naturally-occurring amino acids, including cysteine and cystine, nor with the methionine derivatives homocysteine thiolactone and methionine sulfoxide. The following compounds, which like methionine contain the grouping $-\text{CH}_2-\text{CH}_2-\text{S}-\text{CH}_2-$, gave the reaction in its full intensity: ethionine, homomethionine, hexomethionine and homodjenkolic acid. Methyl-S-cysteine and ethyl thioglycollic acid gave a positive reaction, but less strongly than methionine; benzyl-S-cysteine and ethyl-S-cysteine gave the reaction faintly, and djenkolic acid very faintly. Carboxymethylcysteine and phenyl-S-cysteine gave negative reactions. Carbohydrates and alkaloids did not give a colour with the reagent, whilst some proteins, presumably containing methionine, gave positive responses. The coloured compound appears to be formed by the combination of 1 molecule of the sulphide with 1 molecule of cupric chloride. With an excess of the latter, green colours tend to be formed, and this reduces the sensitivity of the test. The addition of hydrogen peroxide destroys the colour, and affords a valuable method of confirming a positive result. For qualitative purposes, it is best to allow 1 or 2 drops of 0.1 *M* cupric chloride solution in hydrochloric acid to run down the side of a test-tube upon which a few particles of the substance have been scattered. If at the moment of contact an intense brown colour develops, then methionine or a closely related substance is present. F. A. R.

Fluorometric Method for Determining the Riboflavin Content of Foodstuffs. A. Z. Hodson and L. C. Norris. (*J. Biol. Chem.*, 1933, 131, 621-630.)—A 5-g. sample of the material to be tested is weighed into a 500-ml. Erlenmeyer flask, and 50 ml. of 0.25 *N* sulphuric acid are added. After being thoroughly mixed the contents of the flask are gently heated under reflux for an hour and allowed to cool, after which the *pH* is adjusted to 7.0 to 7.5 with trisodium phosphate solution. The extract is diluted to 50 ml. with water and filtered after standing for 30 minutes. An aliquot part of the filtrate is pipetted into a 200-ml. volumetric flask and diluted to approximately 175 ml. with water. Then 2 ml. of sodium hydrosulphite solution (1 g. of pure sodium hydrosulphite and 1 g. of sodium bicarbonate in 20 ml. of ice-cold water kept in an ice-bath; the solution is stable for about 4 hours) and 2 ml. of stannous chloride solution (10 g. of stannous chloride in 25 ml. of conc. hydrochloric acid are diluted 200-fold with water) are added, and the solution is diluted to the mark. The mixture is allowed to stand for 10 minutes to ensure as complete reduction as possible of interfering pigments and fluorescent substances; it is then transferred to a 1-litre Erlenmeyer flask and shaken vigorously for 5 minutes with access to air. The riboflavin present, which is reduced to a non-fluorescent form together with the other substances, is re-oxidised by this procedure, whereas none of the interfering substances so far encountered is thereby affected. A definite volume of the solution is then introduced into the cell of a fluorometer and the fluorescence is

measured in the usual way. In addition, three other readings are taken: (a) with the unknown solution after addition of a known amount of standard riboflavin solution, (b) with the unknown solution after reduction with sodium hydrosulphite solution (2 per cent. of its volume), and (c) with a solution containing the same amount of riboflavin as that added to the unknown. The difference between the apparent riboflavin-contents before and after reduction gives the true riboflavin-content, except in so far as this is affected by the presence of stable interfering pigments. This effect is corrected for by multiplying by the factor:

$$\frac{\text{riboflavin content of the standard solution}}{\text{difference between the riboflavin contents before and after the addition of flavin to the unknown}}$$

The appropriate dilution factors must, of course, be employed where solutions of sodium hydrosulphite or flavin have been added to the unknown. In the extraction of bulky materials, such as alfalfa meals, the quantity of 0.25 *N* sulphuric acid should be doubled. Samples of milk by-products containing casein are preferably extracted with a mixture of acetone (3 volumes) and *N* sulphuric acid (1 volume) rather than 0.25 *N* sulphuric acid. Satisfactory results cannot be obtained with extracts containing large amounts of light-absorbing impurities, as, for instance, those obtained from molasses, but by means of the following modification most of the interfering substances are removed. To 10 ml. of the neutralised extract are slowly added 90 ml. of methyl alcohol and the mixture is filtered, with precautions to prevent evaporation. A suitable aliquot part of the filtrate is transferred to a 200-ml. volumetric flask and diluted to 175 ml. Subsequently the usual procedure is followed. The validity of the results obtained by this method should always be checked by determining the recovery of added riboflavin, since some flavin may be lost on the methyl alcohol precipitate. The riboflavin-contents found by the method were in close agreement with those obtained by a microbiological method, although variations up to 10 per cent. were observed in duplicate estimations. The following are some of the more important results, the riboflavin contents being expressed in γ per ml.: dried alfalfa meal 15, white fish meal 10, liver meal 80, dried skim-milk 20, dried whey 20, dried yeast 30 to 50, wheat, yellow corn and oats (various forms) 0.2 to 1. F. A. R.

Vitamin E Chemistry: Oxidation Products of α -Tocopherol and of Related 6-Hydroxychromans. L. I. Smith, W. B. Irwin and H. E. Ungnade. (*J. Amer. Chem. Soc.*, 1939, **61**, 2424-2429.)—When α -tocopherol, 2,2,5,7,8-pentamethyl-6-hydroxychroman and similar substances are oxidised with ferric chloride, gold chloride, or, in certain circumstances, silver nitrate, the products are yellow *p*-quinones. When the action of silver nitrate is prolonged beyond this stage brilliant red solutions are obtained (John, Dietzel and Emte, *Z. physiol. Chem.*, 1939, **257**, 173; Karrer, Fritzsche and Escher, *Helv. Chim. Acta*, 1939, **22**, 661). Red solutions result also when nitric acid is the oxidising agent as in the photometric analysis of tocopherols (Furter and Meyer, *Helv. Chim. Acta*, 1939, **22**, 240; Abst., *ANALYST*, 1939, **64**, 217). The red crystalline substance (m.p. 109 to 110° C.) present in these solutions has now been obtained from 2,2,5,7,8-pentamethyl-6-hydroxychroman with either silver nitrate or nitric acid as oxidising

agent, and it is shown by comparison of the molecular extinction curves that it is responsible for the colour in the Furter and Meyer reaction. John, *et al.* (*loc. cit.*) showed that the red compounds differ from the *p*-quinones by 1 carbon atom and 4 hydrogen atoms, and Karrer *et al.* (*loc. cit.*) found a similar change when the quinone from 2,5,7,8-tetramethyl-hydroxychroman was converted into the analogous red compound $C_{12}H_{14}O_3$ (m.p. $140^\circ C.$). These authors proposed a tentative structure for this compound, and by analogy the red compound derived from the pentamethyl-chroman would have a *p*-quinonoid structure. It may be deduced from work on the colour of naphthaquinone derivatives (Hooker, *et al.*, *J. Amer. Chem. Soc.*, 1936, **58**, 1163, 1168, 1179, 1181, 1198, 1202) that it is an *o*-quinone, and this is now confirmed by comparison of its absorption spectra curves with those of substances with an *o*-quinonoid structure.

Cook, Macbeth and Winzor (*J. Chem. Soc.*, 1939, 878) have shown that a maximum in the absorption curve at 400 to $440m\mu$ is characteristic of *o*-quinones and does not occur with *p*-quinones. The fact that the red oxidation products show a maximum absorption around $480m\mu$ is considered good evidence of their *o*-quinonoid structure. This is confirmed by their chemical behaviour. The red substance (m.p. 109° to $110^\circ C.$) forms a phenazine (m.p. 151° to $152^\circ C.$) with *o*-phenylenediamine and a tetramethyl phenazine (m.p. 204° to $205^\circ C.$) with tetramethyl-*o*-phenylenediamine. These phenazines are yellow with a pale green fluorescence in alcoholic solution. α , β , and γ Tocopherols give deep red solutions when oxidised in alcoholic solution with nitric acid. It was not found possible to crystallise these oily compounds, but that from α -tocopherol gave a phenazine showing the greenish fluorescence previously mentioned. Formation of the red *o*-quinones is not limited to 6-hydroxychromans. According to Furter (private communication) and Smith, Irwin and Ungnade (*loc. cit.*) the reaction occurs with 5-hydroxycoumarans. Apparently the presence of an alcohol (or possibly a phenol) is essential for the production of the red compound, and experiments showed that the intensity of colour developed in a given time decreased according as the alcohol was primary, secondary or tertiary. Red solutions are not formed in petroleum spirit, acetic acid or acetone, although a mixture of alcohol and acetone may be used with the tocopherols. There is definite evidence that the alcohol is oxidised during the reaction, and that the oxidation is not due to the nitric acid.

A. O. J.

Vitamin A Content of "Light White" Casein. M. K. Maitra and T. Moore. (*Biochem. J.*, 1939, **33**, 1648–1651.)—"Light white" casein (sodium caseinate) was extracted with various solvents, and the extracts were examined colorimetrically for carotene and vitamin A. Hot alcohol extracted fat amounting to 2 per cent. of the casein; the extract was coloured yellow and gave a blue colour when treated with antimony trichloride solution. The amount of carotene and vitamin A calculated to be present, corresponded to a biological activity of 1 i.u. per g. of casein. This was consistent with the activity found by biological test. Ether and ethylene dichloride extracted only a portion of the fat and of the vitamin A-active material.

F. A. R.

Curative Factor (Vitamin H) for Egg White Injury with Particular Reference to its Presence in Different Foodstuffs and in Yeast. P. György. (*J. Biol. Chem.*, 1939, 131, 733-744.)—Rats, and some other animals, develop a skin disorder when fed on a diet containing a high proportion of egg white. The egg white is deprived of its toxic properties by continued heat treatment or by digestion with pepsin or hydrochloric acid. The toxic effect is also neutralised by an organic protective substance, present in different foodstuffs, to which Boas gave the name "protective factor X" and György the name "vitamin H". A large variety of foodstuffs have now been examined for this detoxifying factor, and its main sources are shown to be liver, kidney, yeast and, to a less extent, cows' milk. The factor is absent from concentrates of the pernicious anaemia factor from liver, but present in the residues. It is insoluble in water and in fat, but is liberated in a water-soluble form by autolysis of yeast in presence of toluene. Chloroform reversibly inhibited this apparent enzyme reaction. F. A. R.

Attempts to Isolate the Factor (Vitamin H) Curative of Egg White Injury. P. György, R. Kuhn and E. Lederer. (*J. Biol. Chem.*, 1939, 131, 745-759.)—Although vitamin H is obtained in a water-soluble form by autolysis of yeast (*cf.* preceding abstract) it cannot be obtained by autolysis of liver, nor do the enzymes of yeast liberate the factor. This can be accomplished, however, by digesting the liver with pepsin or, better, with papain, or by heating with sulphuric or hydrochloric acid under pressure. The simplest method of obtaining good yields of vitamin H was by autoclaving liver powder with water for 1 to 2 hours at 200° C. Fresh liver and kidney did not give the vitamin unless acid was present. The factor was concentrated by a series of processes, including precipitation with acetone or alcohol, which removed large amounts of inert material, precipitation with lead acetate, which also removed impurities, adsorption on charcoal followed by elution with a mixture of pyridine, methyl alcohol and water, and treatment with phosphotungstic acid or gold chloride solution, which precipitated the vitamin along with other substances. The last two reagents did not precipitate the vitamin from solutions relatively free from impurities. Vitamin H was irreversibly inactivated by benzoylation, by treatment with formaldehyde, nitrous acid and ketene, and by oxidation with hydrogen peroxide. F. A. R.

Physicochemical Properties of the Factor (Vitamin H) Curative of Egg White Injury. T. W. Birch and P. György. (*J. Biol. Chem.*, 1939, 131, 761-766.)—A solution of vitamin H was distributed equally in the ten compartments of an electrodialysis apparatus, and the pH and biological activity of the solutions at the end of electrodialysis, carried out over a period of 20 to 48 hours, were determined. The experiment indicated that the factor is an ampholyte with an isoelectric point between pH 3 and 3.5. No appreciable loss of potency occurred during the operation, but a five-fold concentration was obtained in the cells to which the active principle migrated. In spite of its acidic properties, vitamin H could not be precipitated by alkaloids. Further concentration was effected by precipitating the barium salt in alcohol at pH 5 or the calcium salt at pH 8. The most active fraction obtained was free from sulphur and phosphorus.

F. A. R.

Photometric Determination of Oestrogens. I. Modified Kober Reaction for Determining the Total Oestrogens in a Mixture of Oestrogenic Steroids. C. Bachman. (*J. Biol. Chem.*, 1939, 131, 455-462.)—Kober (*Biochem. Z.*, 1931, 239, 209) devised a two-stage reaction for the identification of natural oestrogens. This consisted first in heating the oestrogen at 100° C. with phenolsulphonic acid, whereby a yellowish-brown colour was produced. The solution was then cooled, diluted and reheated, whereupon a characteristic pink colour developed. Besides the disadvantage that such a two-stage reaction cannot readily be made quantitative, it was found that the speed of the reaction was different for different oestrogens. Both these defects have been overcome by diluting the phenolsulphonic acid reagent with water, and by carrying out the reaction at 150° C. instead of 100° C. The reagent is prepared by mixing pure conc. sulphuric acid (5.6 volumes) and pure dry melted phenol (3.6 volumes), and then diluting the phenolsulphonic acid so formed with half its volume of water. One to 3 ml. of a solution (in alcohol) of the oestrogen, containing 10 to 60 γ , are introduced into a series of test-tubes (200 \times 18 mm.), and the solvent is evaporated in a current of air with the tubes immersed in a boiling water-bath. The residues left after evaporation are dried *in vacuo* for 1 hour. Corresponding volumes of alcohol are similarly treated, to serve as blanks. Into each tube are put 3 ml. of the reagent, and the series (not more than 8 tubes) is immersed, one tube at a time at 15-second intervals, in a bath of chlorinated diphenyl maintained at 150° C. The contents of each tube are stirred for 10 seconds, and again 2 minutes later for 10 seconds, each of the tubes being thus treated in rotation. After 9.5 minutes, the first tube is transferred to an ice-bath, the other tubes following at 15-second intervals. The same stirring technique is followed as with the heated tubes. After 5 minutes in ice, the solutions are made up to 15 ml. with dilute sulphuric acid (3 volumes of conc. acid to 7 volumes of water), and the colour of each is measured in an Evelyn photoelectric photometer, using a filter giving maximum transmission at 520 $m\mu$. It was found that, although the pink colours obtained with the three oestrogens—oestradiol, oestrone and oestriol—developed at different rates, the absorptions become equivalent for equal concentrations of all three after heating for 9.5 minutes, thus making it possible to estimate the total content of a mixture of the three substances. The technique of heating and cooling outlined above must be strictly adhered to, and under these conditions, the absorption at 520 $m\mu$ for all concentrations between 10 and 60 γ is linear. Various combinations of oestradiol, oestrone and oestriol were tested, and in the data recorded the variation is not greater than ± 1 per cent. The applicability of the reaction to urine extracts was also verified. F. A. R.

II. New Colour Reaction for Oestriol. C. Bachman. (*Ibid.*, 463-468.)—When oestrogens were heated with a mixture of phosphoric acid and sodium *p*-phenolsulphonate at temperatures exceeding 100° C., a violet-pink colour formed with oestriol, whereas little if any colour developed with other steroids. The reaction has been made the basis of a quantitative method of assaying oestriol in the presence of other oestrogens. Into each of the tubes containing 10 to 60 γ of the oestrogen to be tested, and into accompanying control tubes, are introduced

15 ml. of reagent made by dissolving 2 g. of sodium *p*-phenolsulphonate in 100 ml. of 85 per cent. phosphoric acid. The series of tubes is immersed for 9 minutes in a bath maintained at 150° C., and then for 5 minutes in an ice-bath. The details of this procedure are the same as those described in the preceding paper. The colours of the solutions are measured directly without dilution by means of an Evelyn photoelectric colorimeter, using a filter with maximum transmission at 540*mμ*. α -Oestradiol and oestrone had extinction values of approximately one-third and one-sixth that of oestriol, respectively. Consequently in estimating oestriol in presence of one or other of the other oestrogens, a correction has to be applied by means of the following equation:

$$y = \frac{\epsilon (\text{mixture}) - \epsilon (1\gamma \text{ oestrone}) \times C}{\epsilon (1\gamma \text{ oestriol}) - \epsilon (1\gamma \text{ oestrone})}$$

Where *y* is the amount in γ of oestriol in the mixture, *C* is the concentration in γ of total oestrogen determined by the modified Kober reaction, ϵ (mixture) is the extinction value of the unknown solution measured at 540*mμ*, using the phosphoric acid reaction and ϵ (1 γ oestrone) and ϵ (1 γ oestradiol) are the extinction values observed with 60 γ of the pure oestrogens divided by 60. β -Oestradiol gave a very weak pink tint with a strong greenish fluorescence; equilin, 17-dihydroequilin and both 17-dihydroequilenins gave weak brown or orange-brown tints; dehydroandrosterone and Δ^5 -pregnenol-3-one-20 gave a succession of colours culminating in a pale dirty brown. Androsterone, iso-androsterone, testosterone, aetiocholanol-3(α)-one-20, progesterone, pregnandiol, and sodium pregnandiol glycuronide yielded weak yellow or yellowish-brown solutions. Diethyl stilboestriol gave a weak brown colour. With oestriol results agreeing within 2 per cent. of the theoretical value were obtained, whilst oestriol was estimated with a similar degree of accuracy in mixtures of oestriol and oestrone in varying proportions.

F. A. R.

Root-forming Substances used for Propagation Purposes. M. A. H. Tincker and C. H. Unwin. (*J. Royal Hort. Soc.*, 1939, 64, 554–566.)—A further series of results is reported on the effect of 9 root-forming substances on cuttings of various plants, some being species that cannot normally be propagated by means of cuttings. The most generally valuable substances are indolyl butyric acid, 1:2:3:4-tetrahydronaphthylidene acetic acid (m.p. 92° C.) and a mixture of the latter with 3:4-dihydro-1-naphthylacetic acid. The first of these gave the best results with many species, but an equal number of other species gave better results with the second. The indolyl butyric acid produced a more fibrous root system, whilst tetrahydronaphthylidene acetic acid caused thicker roots to be formed, and is probably the most powerful agent causing cell division and root formation. A second group of very useful compounds comprised α -naphthyl acetic acid, 3:4-dihydro-1-naphthylacetic acid and 1:2:3:4-tetrahydro-1-naphthylacetic acid, and a third group 1:2:3:4-tetrahydronaphthylidene acetic acid (m.p. 163° C.) and indolylacetic acid.

F. A. R.

Bacteriological

Effectiveness of Heat Penetration in the Canning of Meat in the Home by the Pressure Cooker. C. I. Nelson and D. Berrigan. (*J. Agric. Res.*, 1939, 59, 465-474.)—A 12-quart home-type of pressure cooker was used in canning solid meat packs of various types, and a group of organisms such as are likely to occur in canned meats was chosen to test the effectiveness of the sterilising process: *Clostridium botulinum*, *Escherichia coli*, a heat-resistant strain of *Streptococcus faecalis* isolated from a can of spoiled meat, and *Bacillus mesentericus*. The variables experimented with were type of pack; steam pressure (10 lbs. and 15 lbs.); can size; duration of process, and methods of cooling. In general, for solid meat packs temperatures reached under 10 lbs. steam pressure were found insufficient to destroy the bacteria tested, and 65 minutes at 15 lbs. pressure (121° C.) appeared to be the minimum time and temperature that could safely be used to allow for the elimination of food-poisoning organisms in the tests on No. 2 cans; 90 minutes for No. 2½ cans, and 110 minutes for No. 3 cans. Cooling was best effected by cold dipping after removal of the cans from the retort. It was found, however, that temperatures attained at 15 lbs. pressure were not uniformly effective in destroying thermoduric organisms, such as *Bacillus mesentericus* and *Streptococcus faecalis*. Heat penetration occurred most readily where least interference with convection currents was met with, *i.e.* in the following order: liquid, chunk meat, ground-meat patties, ground-meat solid pack, and solid chunk. The surprisingly low ratio of efficiency is due to the lag in the heat curves, the gradient of the non-effective part of the curve varying in the order of the packs given above.

D. G. H.

Toxicological

Toxicology of Sodium and Potassium Permanganates. P. Cheramy and A. Lemos. (*J. Pharm. Chim.*, 1939, 30, 249-252.)—It has been shown by Lemos (*J. Pharm. Chim.*, 1939, 30, 206-212; Abst., ANALYST, 1940, 65, 63) that subcutaneous injection of a considerable amount of potassium permanganate into a rabbit produces an abscess which causes the death of the animal. The permanganate could not be administered with the food. Three intra-stomachic injections of potassium permanganate were made at weekly intervals into a rabbit, the amounts administered being 20, 40 and 40 ml. of 5, 5 and 10 per cent. strength. Water only was administered for 36 hours before each dose. Death occurred after the third dose, the animal having been almost unable to absorb nourishment during the three weeks. The stomach, which still contained permanganate, showed important lesions, and was strongly coloured and almost perforated, the kidneys showed signs of haemorrhage and the liver was congested. The manganese contents (in mg. per 100 g.) of various organs were:—for sodium (A) and potassium (B) permanganate respectively: surrenals, 9.5 and 36.6; muscles, 8.0 and 1.2; lung, 6.5 and 5.6; marrow, 5.0 and 25.0; spleen, 4.4 and 21.2; bile, 3.1 and 45.5; brain, 1.5 and 4.0; genital organs, 1.2 and 3.4; blood, 1.1 and —; heart, 1.0 and 3.2; kidneys, 0.70 and 2.3; liver, 0.07 and 6.2. During treatment urea in the blood rose from (A) 0.40 to 0.65 g. per 1000 (when the animal was killed), and (B) from 0.50

to 1.42 g. per 1000 (after the second dose), indicating a lesion of the kidneys. In a case of human poisoning from manganese, in which the manganese-content of the blood was 2.25 mg. per 100 g., the urea in the blood of an anuric invalid was 3.50 g. per 1000. The distribution of the manganese in the rabbit treated was similar to that observed in acute cases of human manganese poisoning. It has been suggested that the toxicity of large doses, especially of the potassium salt, is due to the caustic action of the product, which causes fatal lesions. E. B. D.

Toxicity of Coal Tar Naphtha Distillates. H. Taylor. (*Chem. and Ind.*, 1939, 58, 1078–1080.)—Coal tar naphtha distillates as used for fumigation against bed bugs, are complex mixtures of variable composition with a boiling range of 160–190° C., which may account for the disagreement in results obtained in the present investigation from those of Cameron, Paterson, de Saram and Thomas (*J. Path. and Bacter.*, 1938, 46, 1, 95). Their vapours were found to have considerable narcotic action on rats at concentrations over 0.1 per cent. by volume in air, and in one instance exposure to a 0.16 per cent. concentration caused death in all the animals used. The narcotic effects were very similar for all samples, although these differed greatly in chemical composition, but the effects on the internal organs were widely different. Of the four samples used, two were capable of producing degenerative changes in the liver, and there were also suggestions of kidney damage. The determinations of the concentrations of vapour were made by means of the interferometer, by which any change in concentration is readily detected. It is suggested that possible hazards should be kept in mind in using this method of fumigation. D. G. H.

Water

Volumetric Determination of Sulphate in Water. J. Courtois and P. Bonjean. (*Ann. chim. anal.*, 1939, 21, 229–235.)—The process studied involves the reaction of a soluble sulphate with barium chromate yielding barium sulphate and a soluble chromate which is determined iodimetrically. A method based on this principle and described in "*Einheitsverfahren der physikalischen und chemischen Wasseruntersuchung*" (Berlin, 1936) was tested and found to give low results, owing to adsorption of chromate on the barium sulphate. A modification of this method in which the adsorption effect is minimised is now proposed. A 100-ml. sample of water is heated to boiling and 20 ml. of a solution containing 5 g. of barium chromate and 12.5 g. of hydrogen chloride per litre are added slowly. The boiling is maintained for 5 minutes, after which the liquid is allowed to cool for 15 minutes in order to secure complete precipitation of barium sulphate. The liquid is again boiled, ammonia is added, drop by drop, until the colour changes from reddish to greenish; and the boiling is continued for 5 minutes. This precipitates the barium chromate, leaving soluble chromate in solution proportionate to the sulphate originally present. The liquid is cooled, diluted to 200 ml. and filtered. To a 100-ml. portion of the filtrate 5 ml. of 25 per cent. sulphuric acid (by volume) and 5 ml. of a freshly prepared 30 per cent. potassium iodide solution are added, and after a 10-minute interval the liberated iodine is titrated with *N*/100 thio-sulphate solution, with starch as indicator. It is necessary to make a correction

for the solubility of barium chromate, amounting to 0.85 ml. of $N/100$ thiosulphate solution to be deducted from the titration value of 100 ml. of test solution. Test-results obtained with solutions containing 38 to 510 mg. of SO_4 per 100 ml. were all consistently about 3.5 per cent. low, suggesting that it would be desirable to augment results by this amount. S. G. C.

Agricultural

Determination of Hydrocyanic Acid by the Picric Acid Method and the KWSZ Photometer. J. T. Sullivan. (*J. Assoc. Off. Agr. Chem.*, 1939, **22**, 781-784.)—The picric acid test for hydrocyanic acid (Guignard, *Compt. rend.*, 1906, **142**, 545) is commonly used for qualitative purposes and has recently been applied to the quantitative determination of hydrocyanic acid in plant products (Rogers and Frykholm, *J. Agr. Res.*, 1937, **55**, 533; Boyd *et al.*, *J. Amer. Soc. Agron.*, 1938, **30**, 569; Doak, *New Zealand J. Sci. Tech.*, 1938, **10**, 163). The procedure here described is an adaptation of Boyd's method to the determination of hydrocyanic acid in individual white clover plants. Owing to the limited amount of material available a determination of less than 0.05 mg. of hydrocyanic acid was often necessary. Such small quantities could be more accurately determined by the KWSZ photometer than by visual comparison with standards. Fresh white clover leaves (10 g.) were allowed to stand with 5 ml. of toluene for several days at room temperature in a rubber-stoppered 500-ml. short-necked Kjeldahl flask. The mixture was steam-distilled from the same flask into 5 ml. of 2 per cent. potassium hydroxide solution until 80 to 90 ml. of distillate had been collected. The distillate was diluted to 100 ml. with water, the supernatant toluene was removed, and 20 ml. or less were treated with alkaline picrate solution (25 g. of anhydrous sodium carbonate and 5 g. of dry picric acid in 1 litre) in a test-tube, and if less than 20 ml. of the original solution had been taken the volume was made up to 30 ml. with water. The test-tube, loosely plugged with cotton wool, was heated in boiling water for exactly 5 minutes. A control tube containing water and alkaline picrate solution was heated simultaneously in the same manner. The contents of the sample tube were placed in one cell of the photometer and the control liquid in the other. A solution of 10 g. of crystalline copper sulphate and 1 drop of conc. sulphuric acid in 1 litre of water was used as the light filter, and the transmission of the sample was compared with that of the control liquid. A transmission curve for the instrument was constructed by means of a standard solution of potassium cyanide containing 0.4 mg. of hydrocyanic acid per ml. From a ten-fold dilution of this solution a series of standards was made by diluting to 100 ml. portions ranging from 1.25 to 25.0 ml. These standard solutions (20 ml.) were used to construct the calibration curve of the instrument. It was found inadvisable to use solutions containing more than 0.2 mg. of hydrocyanic acid. The conditions under which hydrocyanic acid is liberated and distilled from its particular glycoside needs further study especially if this method is to be applied to plants other than white clover. In most instances the period of autolysis before distillation caused the liberation of more hydrocyanic acid than the amount liberated by immediate distillation. Omission of the preservative in the

autolysis, or freezing and thawing followed by distillation with or without a preservative, led to variable results. Immediate heating of the distillate with alkaline picrate solution is necessary, but the examination in the photometer may be delayed for a day or two. In order to compare results obtained by this process with those of the alkaline titration method (*J. Assoc. Off. Agr. Chem.*, 1936, **19**, 94) it was necessary to take larger samples and to use large dilutions for the picric acid test. Under these conditions the picric acid method gave results 8 per cent. higher on an average than those obtained by the alkaline titration method.

A. O. J.

Determination of Boron in Soils and Plants by means of Quinalizarin.

K. C. Berger and E. Truog. (*Ind. Eng. Chem., Anal. Ed.*, 1939, **11**, 540-545.)—The colour reaction is most sensitive in a medium of 93 per cent. by weight of sulphuric acid (*cf.* Smith, *ANALYST*, 1935, **60**, 735). Colour standards are prepared by diluting measured volumes of dilute boric acid solution in boron-free test-tubes to 1 ml. with water, adding 9 ml. of 98.5 per cent. sulphuric acid, cooling, and adding 0.5 ml. of quinalizarin solution (0.01 g. in a mixture of 90 ml. of 98.5 per cent. sulphuric acid with 10 ml. of water); the colour develops fully in 15 minutes and remains permanent in stoppered tubes. The use of standards covering the range 0.001 to 0.01 mg. of boron is suggested. Available boron in soils is determined by extraction of a 20-g. sample with 40 ml. of water by boiling under a reflux condenser for 5 minutes. An aliquot portion of the clear extract is evaporated to dryness with 0.1 g. of potassium carbonate in a platinum dish; the residue is ignited, and, after cooling, triturated with 5 ml. of 0.4 *N* sulphuric acid. To a 1-ml. portion of the solution are added 9 ml. of 98.5 per cent. sulphuric acid, the mixture is cooled, 0.5 ml. of quinalizarine reagent is added, and the colorimetric comparison is made after 15 minutes. Total boron in soils may be determined by fusing a 0.5 g. sample with 3 g. of sodium carbonate in a platinum crucible. The melt is dissolved in water and acidified with sulphuric acid to a *pH* value of 5.5 to 6.0; 350 ml. of alcohol are added, and the liquid is diluted to 500 ml. and filtered. A 400-ml. portion of the filtrate is made alkaline with potassium carbonate and evaporated to dryness; the residue is ignited and triturated with 5 ml. of 0.4 *N* sulphuric acid, and the boron is determined as described above. For the determination of the total boron in plants, the plant tissue is ignited to a grey ash, which is extracted with dilute sulphuric acid, and the quinalizarin test is applied to the extract.

S. G. C.

Organic

Volumetric Determination of Organic Substances by Chromic Oxidation: Nitro-chromic Solutions. **M. H. Cordebard.** (*J. Pharm. Chim.*, 1939, **30**, 263-272.)—The advantages and disadvantages of oxidation by mixtures of potassium dichromate and (a) conc. sulphuric acid, (b) dilute sulphuric acid (1:1), are discussed. With (b) oxidation of ethyl alcohol to acetic acid is not quantitative, as the acid is partly decomposed slowly into carbon dioxide and water. With a mixture of (b), potassium dichromate and silver nitrate, it has been shown that the presence of free nitric acid does not interfere with the dichromate titrations

(cf. Cordebard, *Bull. Soc. Chim.*, 1928, 43, 97). The use of "nitro-chromic" solutions (i.e. of potassium dichromate in nitric acid) is therefore proposed. Official nitric acid dissolves approximately 150 g. of potassium dichromate per litre at room temperature; standard solutions can therefore be prepared without risk of crystallisation of chromic anhydride. Normal and decinormal solutions are remarkably stable (they can be evaporated in air to about a fourth of their initial volume without appreciable loss of strength) and the oxidising power of nitro-chromic solutions, though not equal to that attainable with (a), is superior to that of sulphuric-chromic solutions of the same temperature and acidity. Most substances completely oxidisable to carbon dioxide and water with (b) are similarly oxidisable with nitro-chromic solutions, e.g. methyl alcohol, formaldehyde, formic acid, glycerol in the cold; tartaric, oxalic and citric acids, sugars (in a few minutes on the boiling water-bath); most organic compounds containing oxygen (at 122° C., the b.p. of the mixture, after different periods). Acetic acid is not partly oxidised to carbon dioxide as it is with (b); hence alcohol can be determined by oxidation to acetic acid, with nitro-chromic solution and subsequent iodimetric determination of the excess of dichromate. Alcohol in aqueous solution is first diluted to (a) 1 per cent. at the maximum or (b) less than 0.2 per cent. Ten ml. of *N*/10 nitro-chromic solution are shaken in a stoppered flask with either 1 ml. of (a) for 2 minutes or 5 ml. of (b) for 5 minutes, 40 ml. of water are added, and the mixture is transferred to a vessel containing 100 ml. of 1 per cent. potassium iodide solution and titrated, after 1 minute, with *N*/10 sodium thiosulphate solution. The greatest care in diluting the alcoholic solution is essential. The following determinations of alcohol in the presence of non-oxidisable substances are described:—(a) *Anaesthetic chloroform* (containing little alcohol):—Ten ml. of *N*/10 nitro-chromic solution are shaken vigorously with 8 ml. of the chloroform for 5 minutes to form a temporary emulsion, 50 ml. of water and 100 ml. of 1 per cent. potassium iodide are added, and the mixture is titrated, after 2 minutes, with *N*/10 sodium thiosulphate solution until the aqueous layer is blue. (b) *Camphorated alcohol or camphorated brandy* (containing about 95 per cent. of alcohol):—Exact preliminary measurement is important; it is made by pipetting 10 ml. of the alcohol into a 1-litre graduated flask and adding water, drop by drop at first, and then more quickly, until the vessel is full. The liquid is filtered or decanted, and the alcohol is determined as before on 1 ml. When other oxidisable substances are present a preliminary separation of the alcohol must be made before oxidation.

E. B. D.

***m*-Nitrobenzazide as a Reagent for the Identification of Phenols.**

P. P. T. Sah and T. F. Woo. (*Rec. Trav. Chim. Pays-Bas*, 1939, 58, 1013–1017.)—In boiling ligroin solution *m*-nitrobenzazide decomposes into gaseous nitrogen and *m*-nitrophenyl isocyanate, the latter reacting with phenols to form crystalline *m*-nitrophenyl urethanes which can be used for the identification of the phenols. The following procedure is recommended:—A mixture of the azide (0.5 g.), or an equivalent quantity of the isocyanate (prepared by refluxing the benzazide in anhydrous toluene solution in absence of moisture, removing the solvent by vacuum distillation and crystallising the residue from ligroin), and a molecular

equivalent of the phenol is heated under reflux for 4 hours with 5 ml. of anhydrous ligroin and allowed to stand overnight. If necessary, the solution is then concentrated to a small volume, and the crystals of urethane are filtered off with the aid of suction and examined under the microscope. The yield is weighed and the m.p. is determined before the product is purified by recrystallisation from a suitable solvent. The author tabulates the m.p., crystalline form, solvent used for recrystallisation, formulae, and nitrogen-contents of the urethanes prepared from 33 phenols.

E. M. P.

3.5-Dinitro-4-methylbenzazide as a Reagent for the Identification of Amines. P. P. T. Sah. (*Rec. Trav. Chim. Pays-Bas*, 1939, **58**, 1008–1012.)—Refluxing amines with 3.5-dinitro-4-methylbenzazide in toluene solution leads to the formation of crystalline 3.5-dinitro-4-methylphenyl ureas which can be used for the identification of the amines. Of the amines examined, the primary amines gave the best results; the secondary amines yielded products which were relatively more soluble in benzene or toluene but which could be isolated by concentrating the solution or by using ligroin as the solvent. The amides also reacted, but the yields were much less. The procedure adopted was as follows:—A mixture of the azide (0.25 g.) and a molecular equivalent of the amine was dissolved in 5 to 10 ml. of anhydrous toluene, and the solution was heated under reflux on the sand-bath for 4 hours. After standing overnight in the cold the crystalline precipitate was filtered off and weighed. The melting-point was determined and the crystalline form examined under the microscope, after which the product was purified by recrystallisation from a suitable solvent (acetone, ethyl acetate, benzene, ligroin, 95 per cent. ethyl alcohol, methyl-ethyl ketone, or a mixed solvent). The author tabulates the m.p., the crystalline form, the solvent used for recrystallisation, the formula and the nitrogen-content of the 3.5-dinitro-4-methylphenyl ureas prepared from 50 amines.

E. M. P.

Phthalisation in the Presence of Pyridine. S. Sabetay. (*Ann. Chim. anal.*, 1939, **21**, 289–290.)—The method previously described (*Ann. Chim. anal.*, 1937, **19**, 285; *Abst.*, *ANALYST*, 1938, **63**, 61) for the determination of primary and certain secondary alcohols by phthalisation, *i.e.* heating with phthalic anhydride in the presence of pyridine and subsequent conversion of the excess of phthalic anhydride into phthalic acid which is determined by titration, does not give concordant results unless there is rigorous adherence to the specified conditions. The instability of the acid phthalic esters renders this necessary, especially as both aqueous pyridine and phthalic acid exert a saponifying action on them. In an investigation of the phthalisation of benzyl alcohol, the effect of prolonging the duration of heating and of varying the amount of water added for hydration was studied. Both these variations were found to affect the process adversely, and it is recommended that the following procedure should be followed minutely. A quantity of the sample (*e.g.* essential oil) equivalent to 0.5 to 1.0 g. of alcohol is treated in an acetylation apparatus, fitted with accurately ground-in glass joints, with 10 ml. of the phthalic anhydride and pyridine mixture (*loc. cit.*). The amount of sample to be taken is such that it represents about four times the amount of phthalic anhydride theoretically necessary for the reaction. A control

experiment is made simultaneously. Both acetylation flasks are heated in boiling water, and at the end of one hour exactly 50 ml. of water are added to each. Within one minute the flasks are removed from the bath and cooled rapidly, and their contents are titrated with alcoholic *N*/2 potassium hydroxide solution in presence of phenolphthalein. The subsequent procedure is as previously described (*loc. cit.*).

A. O. J.

Glasses: Organic and Inorganic. H. Moore. (*Chem. and Ind.*, 1939, 58, 1027-1037.)—At the International Congress on Glass in London, 1936, Eitel and Weyl proposed the following classification of glasses:—(1) Inorganic glasses: derived from silicates, borates, etc. (2) Organic glasses: yielded by undercooled organic substances and showing reversible softening behaviour. (3) Transparent, glass-clear synthetic products obtained not by the undercooling of fusions, but by the condensation or polymerisation of simple constituents. Organic substances with claims to be considered as potential glass substitutes may be grouped as follows:—Group A.—Cast phenol formaldehyde resin, moulded transparent phenol formaldehyde resin, moulded transparent urea-formaldehyde resin, moulded colourless alkyd resin. Group B.—Polystyrene, polymethylmethacrylate and other methacrylic esters, polymethylacrylate, polymethacrylic nitrile, polyvinylmethyl ketone, polyvinylphenyl ketone, polyisopropenyl methyl ketone, polyvinyl acetate, polyvinyl chloride, polyvinyl acetals and co-polymers of many of these substances. Group C.—Cross-linked polymers. Group D.—Regenerated cellulose, cellulose acetate, cellulose nitrate, cellulose acetobutyrate, ethyl and benzyl cellulose, chlorinated deproteinised rubber. The structure and properties of the different organic glasses are discussed, and a description is given of their physical properties as compared with those of inorganic glasses. The following table, largely compiled from published data, summarises some of the physical properties of transparent resins compared with those of glass:

| | Cellulose acetate | Cellulose nitrate | Polymethyl methacrylate | Polystyrene | Polyiso- propenyl methyl ketone |
|---|------------------------------|---------------------------|----------------------------|------------------------------------|--|
| Sp.gr. | 1.27-1.37 | 1.35-1.60 | 1.18 | 1.05-1.07 | 1.11-1.15 |
| Spec. heat (cal. per °C. per g.) | 0.31-0.39 | 0.34-0.38 | 0.45 | 0.324 | — |
| Softening pt. (°C.) . . . | 60-71 | 71-91 | 77-113 | 104-116 | 60-80 |
| Tensile strength (lb. per sq. in.) | 4000-9100 | 5000-10,000 | 4000-6000 | 5000-5500 | — |
| Volume resistivity (ohm per cm.) | (4.2-6.2) × 10 ¹³ | (2-30) × 10 ¹⁰ | 10 ¹⁵ | 10 ¹⁷ -10 ¹⁸ | — |
| Modulus of elasticity (lb. per sq. in. × 10 ⁵) . . . | 2.0-3.5 | 2.0-4.0 | 6.0 | 4.0-6.0 | — |
| Refr. index, <i>n</i> _D | 1.47-1.50 | 1.50 | 1.50-1.52 | 1.60-1.67 | 1.52 |
| Water absorption, per cent (48 hours) | 2.6-3.1 | 1.0-3.0 | 0.5 | 0.05 | 0.3 |
| Brinell hardness | 8.6-12.2 | 8-11 | 18-20 | 20-30 | 20 |
| Power factor (10 ⁵ cycles) | 0.035-0.06 | 0.07-0.10 | 0.02 | less than 0.0002 | |
| Thermal conductivity (10 ⁻⁴ cal. per sec. per sq. cm. per 1° C. per cm.) . . | 4.5-7.6 | 3.1-5.1 | 4.3-6.8 | 1.9 | |
| Dielectric constant (10 ⁵ cycles) | 4.0-5.0 | 6.15 | 2.8 | 2.6 | |
| Thermal expansion (10 ⁻⁵ per °C.) | 14-16 | 12-16 | 7-9 | 6.5-7.5 | |

| | Polyvinyl chloride acetate | Ethyl cellulose | Urea- formal- dehyde | Moulded transparent phenol formal- dehyde | Cast transparent phenol formal- dehyde |
|---|----------------------------------|-----------------------|----------------------------|---|--|
| Sp.gr. | 1.34-1.36 | 1.14 | 1.48 | 1.28 | 1.27-1.32 |
| Spec. heat (cal. per °C. per g.) | 0.24 | 0.25-0.40 | — | — | 0.3-0.4 |
| Softening pt. (°C.) . . . | 55-72 | 99-130 | none | none | none |
| Tensile strength (lb. per sq. in.) | 8000-10,000 | 7000-9000 | 4000-6000 | 6000-8500 | 5000-12,000 |
| Volume resistivity (ohm per cm.) | over 10^{14} | 10^{15} | $(2-2.8) \times 10^{13}$ | 7.5×10^8 | 10^9-10^{14} |
| Modulus of elasticity (lb. per sq. in. $\times 10^6$) . . . | 3.5-4.1 | 2.0-4.0 | 16 | 7-10 | 5-15 |
| Refr. index, n_D | 1.53 | 1.47 | 1.54-1.6 | — | 1.5-1.7 |
| Water absorption, per cent. | 0.05-0.15 (24 hours) | 1.25 (48 hours) | 1.0-2.0 (24 hours) | 0.1 (48 hours) | 0.01-0.5 (24 hours) |
| Brinell hardness | 15-25 | 10 | 48-54 | — | 30-45 |
| Power factor (10^6 cycles) . . | 0.018 | 0.007-0.03 | 0.01-0.03 | 0.019 | 0.01-0.045 |
| Thermal conductivity (10^{-4} cal. per sec. per sq. cm. per 1° C. per cm.) . . . | 4.0 | 5.6 | 7.1 | — | 3.5 |
| Dielectric constant (10^6 cycles) | 4.0 | 2.0-3.0 | 6.0 | 4.5 | 5-7 |
| Thermal expansion (10^{-5} per °C.) | 6.9 | 10-14 | 1.5 | — | 2.8 |
| | Sheet window glass | Soft soda glass | Lead flint glass | Boro- silicate glass (Pyrex) | Fused silica (Vitreosil) |
| Sp.gr. | 2.50 | 2.45 | 4.0 | 2.25 | 2.21 |
| Spec. heat (cal. per °C. per g.) | — | — | 0.12 | 0.20 | 0.20 |
| Softening pt. (°C.) | 600 | 570 | 400 | 600 | 1300 |
| Tensile strength (lb. per sq. in.) | 7000 | — | 7000-17,000 | 7000-14,000 | 4000 |
| Volume resistivity (ohm per cm.) | 20×10^{12} | 5×10^{11} | — | 10^{14} | 5×10^{12} |
| Modulus of elasticity (lb. per sq. in. $\times 10^6$) . . . | 110 | 82 | — | — | 101 |
| Refr. index, n_D | 1.51 | 1.48 | 1.8 | 1.47 | 1.45 |
| Water absorption, per cent. | — | — | — | — | — |
| Brinell hardness | 380 | — | 160-350 | — | 405 |
| Power factor (10^6 cycles) . . | — | — | — | 0.0028 | less than 0.001 |
| Thermal conductivity (10^{-4} cal. per sec. per sq. cm. per 1° C. per cm.) . . . | — | — | — | — | 25 |
| Dielectric constant (10^6 cycles) | 6-7 | — | 7-10 | 4.48 | 3.7-3.8 |
| Thermal expansion (10^{-5} per °C.) | 0.93 | 1.2 | 0.9 | 0.32 | 0.054 |

Determination of Catechin and Catechin-Tans in Gambir. C. J. van Hulssen and D. R. Koolhaas. (*Rec. Trav. Chim. Pays-Bas*, 1939, 58, 831-838.)—About 1 g. of gambir is dissolved in about 200 ml. of hot water and the insoluble residue is filtered off on a 1 G 4 Jena glass crucible, washed with water, dried at 104° C. and weighed as the "impurity"-content of the sample. The filtrate and wash water are combined and made up to 250 ml. Twenty-five ml. of the filtrate are treated with 25 ml. of Stiasny's formalin-hydrochloric acid reagent (100 ml of 35 per cent. hydrochloric acid, 50 ml. of water and 200 ml. of 30 per cent. formalin) and, after shaking, are allowed to stand for 24 hours at room temperature. The precipitate is separated on a 1 G 4 Jena crucible, washed with 100 ml. of

water, dried at 104° C. and weighed. It consists of a combination of the formalin compounds of catechin and of the tanning agents. A further 25 ml. of the filtrate is treated with 25 ml. of gelatin and sodium chloride solution (25 g. of gelatin dissolved in 875 ml. of water, then saturated with sodium chloride, and filtered), and afterwards with 50 ml. of acid sodium chloride solution (saturated sodium chloride solution containing 25 ml. of conc. sulphuric acid per litre), with shaking. After some time 1 g. of kaolin is added, and the mixture is well shaken and filtered. Twenty-five ml. of formalin and hydrochloric acid reagent are added to 50 ml. of the filtrate, and the weight of the precipitate, which is the formalin compound of catechin, is determined. The conversion factors for tanning agents and for catechin were found to be 0.96 and 0.94 respectively, and the results are calculated as follows:—

$$\begin{aligned}
 &\text{Weight of (catechin + tanning agent) formalin compound from} \\
 &\quad 25 \text{ ml. of extract} \quad \quad \quad = a \text{ g.} \\
 &\therefore \text{weight in total extract} \quad \quad = 10a \text{ g.} \\
 &\text{Weight of catechin-formalin compound (after treatment with} \\
 &\quad \text{gelatin) in 50 ml.} \quad \quad \quad = b \text{ g.} \\
 &\therefore \text{weight in 250 ml.} = 2 \times 10 \times b \quad = 20b \text{ g.} \\
 &\therefore \text{weight of tanning agent} \quad \quad = (10a - 20b) \times 0.96 \text{ g.} \\
 &\quad \text{and weight of catechin} \quad \quad = 20b \times 0.94 \text{ g.}
 \end{aligned}$$

E. M. P.

Analysis of Synthetic Resins containing Maleic Acid. E. Sadolin. (*Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 608–610.)—The chemical analysis of maleic acid (anhydride) resins is new, and the method put forward is based on the typical solubility of maleic-abietic acid in faintly acid aqueous solution as contrasted with the constituents of rosin and phenol-formaldehyde resins. Phthalic acid from phthalic resins may be removed in advance. The resin (0.2 g.) is dissolved in 5 ml. of benzene and saponified with 2 ml. of *N* potassium hydroxide in 90 per cent. ethyl alcohol for one hour followed by heating for 30 minutes without a condenser in a steam-bath. To the residue are added 50 ml. of water, and the flask is again heated on the steam-bath for 30 minutes, after which water is added to bring the volume to 200 ml. With methyl red as indicator 6 to 7 ml. of 4 *N* acetic acid are added until the colour approaches red, but the precipitate must settle before the colour is observed. The *pH* is about 4.5. After filtering, washing twice and boiling for 10 minutes (to remove any hydrogen sulphide), precipitation is carried out with 5 ml. of a 1 in 10 lead acetate solution. The flocculated colloidal precipitate is cooled and collected on a Jena glass filtering crucible (10 G 4), the operation taking about 2 hours. The precipitate is dried for 45 minutes at 80° to 90° C. and weighed. The factor for the maleic acid and lead precipitate is approximately 0.30, it being assumed that 60 per cent. of the maleic acid used in the resin is found by this method. On heating the precipitate with dilute hydrochloric acid the maleic-abietic acid separates as a resinous lump, whilst phthalic and similar acids will dissolve. Great care is necessary with the various operations, *e.g.* a double sample of resin precipitated in half the solvent produces a precipitate of about one-third the volume of that described above, and the strength of the

potassium hydroxide solution, time of saponification and of evaporation, etc., profoundly affect results. With the procedure described neither colophony nor the phenol formaldehyde resins give lead precipitates, but phthalic acid resins form an easily distinguishable precipitate. The method gives a quantitative determination of maleic-abietic acid and duplicate tests agree to within 2 per cent. The quantity of the acid diminishes during the production of resin from approximately 10 per cent. to an average of 60 to 70 per cent. It is thus possible to control the process of resin production and to form an idea as to the quantity of maleic acid entering into an unknown resin, and so to aid in its identification. D. G. H.

Inorganic

Determination of Bismuth by Precipitation as Quinaldine Iodobismuthite. J. R. Hayes and G. C. Chandlee. (*Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 531-532.)—To the solution of bismuth as sulphate in dilute sulphuric acid (acidity about 1 *N*; volume 200 ml.) are added 15 ml. of 10 per cent. sodium sulphite solution, and the bismuth is precipitated by the addition, drop by drop, with stirring, of 20 ml. of reagent solution (150 ml. of quinaldine, 50 ml. of conc. sulphuric acid and 75 g. of potassium iodide per litre). The precipitate of quinaldine iodobismuthite is allowed to settle out for about 15 minutes, filtered off on a sintered glass filter, and washed, first with 40 to 50 ml. of a solution of 35 ml. of quinaldine, 15 ml. of conc. sulphuric acid and 0.8 g. of potassium iodide per litre, then with 30 ml. of a 90:10 mixture of dibutyl ether and acetone (water, which decomposes the precipitate, should not be used for washing). The precipitate is decomposed by digestion in boiling 5 per cent. sodium hydroxide solution for 20 minutes. After cooling, the liquid is acidified and the iodide-content of the solution is determined volumetrically by iodate titration as in Lang's iodine-cyanide method; 1 ml. of 0.1 *N* (0.025 *M*) potassium iodate solution is equivalent to 0.002612 g. of bismuth. In test experiments good results were obtained with 0.03 g. of bismuth in presence of amounts of a similar order of the following metals: antimony, lead, cadmium, copper, iron, tin (stannous and stannic), arsenite, arsenate, nickel, chromium, cobalt, manganese, calcium, beryllium, uranyl, aluminium, titanium and barium. Slightly modified precipitation conditions were employed in presence of some ions, thus: antimony, addition of 4 g. of ammonium tartrate before precipitation of bismuth; lead, prior precipitation and filtration of lead sulphate; cadmium, addition of 5 ml. of pyridine to the test solution; phosphate, acidity adjusted to 4 *N* to prevent precipitation of bismuth phosphate. Low results were obtained when appreciable amounts of chloride were present. Silver and mercury interfered. As little as 0.0003 g. of bismuth was determined accurately with the use of 0.01 *N* potassium iodate solution for the titration by Andrews' iodine chloride method. S. G. C.

Permanent Artificial Standards for the Nephelometric Determination of Arsenic with Bougault's Reagent. J. Thuret. (*Ann. Falsif.*, 1939, 32, 328-330.)—The dark coloured turbidity of elemental arsenic given by Bougault's reagent (hypophosphorous acid) slowly subsides in spite of the presence of stabilisers

such as gum arabic. A liquid is proposed which simulates the characteristics of the arsenic turbidity but remains permanent. It is prepared by heating together 150 ml. of water containing 2.5 g. of borax with 1 g. of powdered rosin (colophony) for 15 to 20 minutes, and allowing the emulsion to cool, with continuous shaking throughout. The standards are prepared by matching suitably diluted portions of the emulsion against freshly prepared suspensions of arsenic precipitated by hypophosphorous acid. The standards remained unchanged for 1 month.

S. G. C.

Elimination of Iron by Cupferron Prior to Colorimetric Determination of Lead with Dithizone. L. Panouse-Digeaud and H. Cheftel. (*Ann. Falsif.*, 1939, 32, 296-301.)—Appreciable amounts of iron interfere with the dithizone colorimetric determination of lead, even in presence of potassium cyanide. The authors add to the test solution, rendered distinctly acid with hydrochloric acid, sufficient cupferron to precipitate the iron. The iron-cupferron precipitate is then removed by chloroform, the aqueous portion is evaporated to white fuming with sulphuric and nitric acids in order to destroy the excess of cupferron, and the lead in solution is then determined by the dithizone method in the usual manner.

S. G. C.

Direct Determination of Alumina in Silicates. E. W. Koenig. (*Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 532-534.)—Among silicates considered were felspar, lepidolite, china clay, beryl, cyanite, glass sand, spodumene and a burnt refractory. The aluminium is ultimately precipitated with hydroxyquinoline, residual iron being kept in solution as the ferrous complex with $\alpha\alpha'$ -dipyridyl or *o*-phenanthroline. A sample-weight of the finely ground material sufficient to yield 10 to 30 mg. of alumina is fused with sodium hydroxide in a nickel vessel. The melt is extracted with water, the liquid is boiled, and nickel and ferric hydroxides are filtered off and discarded. To the filtrate containing the aluminium 15 ml. of 8-hydroxyquinoline solution (2.5 per cent., in dilute acetic acid) are added, and hydrochloric acid is introduced with stirring until the precipitate which first forms dissolves completely and the solution is distinctly acid. A greenish colour appearing during this process indicates the presence of ferric iron. Sufficient hydroxylamine hydrochloride to reduce the iron is added, and the solution is heated to 80° to 90° C. An excess of $\alpha\alpha'$ -dipyridyl or of *o*-phenanthroline is added, and the aluminium is precipitated by the hydroxyquinoline present by the addition of ammonium acetate solution (3 per cent. concentration). The suspended hydroxyquinoline complex is caused to coagulate by stirring (not by boiling), and is filtered off and washed with cold water. The precipitate is dissolved in hot dilute hydrochloric acid and the aluminium is determined by bromate-bromide titration of the hydroxyquinoline in the usual way.

S. G. C.

Stability of Calcium Hypochlorites. A. Guillaume and Y. Nicolas. (*Ann. chim. anal.*, 1939, 21, 261-266.)—Ordinary bleaching powder has been compared with French proprietary hypochlorite powders having a considerably higher content of available chlorine, such as Chlorfix, Jav, Perchlorfix and Perfix.

The keeping properties of ordinary bleaching powder were inferior, particularly when the substances were stored in wooden barrels exposed to damp and varying temperature. The stability of the proprietary materials was satisfactory when diluted with other substances; sand, talc and coke cinder were the best for this purpose. Ready-prepared mixtures of *e.g.* Perfix with sand can be kept in metal drums or stoneware vessels for several months without marked deterioration, a point of importance in connection with the use of hypochlorite for decontamination from war gases.

S. G. C.

Composition of Lithium and Potassium Salts Precipitated by Uranyl Acetate Reagents for Sodium. E. R. Caley and W. O. Baker. (*Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 604–607.)—Certain uranyl acetate reagents for sodium also precipitate lithium from concentrated solutions, and the present investigation shows that the lithium precipitates are always triple acetates, analogous to the triple acetates precipitated from sodium solutions by the same reagents. The water-content of these salts when isolated is slightly variable; approximately 6 mols. of water of hydration are present when the salts are dried to constant weight at 100–105° C. The experimental cadmium, mercury and copper uranyl acetate reagents tried gave no precipitates with concentrated lithium solutions. From concentrated solutions potassium also may be precipitated by uranyl acetate reagents for sodium, always as $\text{KUO}_2(\text{C}_2\text{H}_3\text{O}_2)_3$. The copper uranyl acetate reagent used appears to be the most nearly specific qualitative reagent for sodium, since it is moderately sensitive to sodium, yet forms no precipitate with lithium solutions.

D. G. H.

Colorimetric Estimation of Silica and its Use in the Metallurgy of Aluminium. P. Urech. (*Helv. Chim. Acta*, 1939, 22, 1023–1036.)—The formation of a greenish-yellow colour by the interaction of silicates and ammonium molybdate has been applied to the determination of silicon in crude aluminium and recast metal, of silicon in refined aluminium, of silicic acid in clays, and of silica in fluorides. The following methods are recommended:

Silicon in crude aluminium and recast metal.—Sodium hydroxide (2.5 g.) or 15 per cent. sodium hydroxide solution (16.7 g.) is added to 20 ml. of water in a nickel dish of 10 cm. diameter (the sodium hydroxide solution must be stored in a pure nickel flask). One g. of metal filings is added, and the mixture is heated in the dish, which is covered with a pure nickel cover, until reaction begins. After the metal has dissolved the liquid is heated for a short time, cooled, and poured into a 300-ml. beaker containing 50 ml. of 6 *N* nitric acid. The solution is heated until clear, transferred to a 500-ml. graduated flask, diluted to about 450 ml. with hot water (the temperature of the solution should be 70–75° C.), treated with 10 ml. of 10 per cent. ammonium molybdate solution, cooled under running water, and made up to volume. The intensity of colour is measured in a Pulfrich photometer, a 50-mm. layer of solution and an S43 filter being used. The silicon-content of the aluminium is read from a standard curve prepared by applying the same technique to a solution of 0.01 g. of 99 per cent. silicon in 100 ml. of sodium hydroxide solution.

Silicon in refined aluminium.—The method of R. Gadeau (*Ann. Chim. anal.*, 1937, 3, 64) is modified by using hydrochloric acid instead of nitric acid and omitting the heating to 70° C. Standard curves are prepared as follows:—A 15 per cent. solution of sodium hydroxide (33.3 g.) or solid sodium hydroxide (5 g.) and 20 ml. of water are heated to boiling in a nickel dish of 10 cm. diameter, cooled, and poured into a 300-ml. Jena glass flask containing about 80 ml. of water. The solution is treated with 27 ml. of 6 *N* hydrochloric acid, transferred to a 300-ml. graduated flask, cooled to room temperature, treated with 10 ml. of 10 per cent. ammonium molybdate solution, and made up to volume. The flask is shaken and left for 10 minutes, and the colour of the liquid is matched with that of a standard picric acid solution. The colour may also be measured in a Pulfrich photometer. The figure obtained is the zero point of the curve, corresponding with absence of silicon. Other values are obtained by the use of measured quantities of a standard sodium silicate solution, and a curve is drawn.

Determination of silicic acid in clays.—One g. of clay and 5 g. of a 3:1 mixture of anhydrous sodium carbonate and anhydrous borax are mixed with a platinum wire in a platinum crucible, which is covered and heated first over an ordinary burner and then over a blow-lamp until no more carbon dioxide is evolved. The crucible and the melt are heated with water in a nickel dish of about 500-ml. capacity until the melt is completely dissolved, the solution is cooled and filtered, and the filtrate and washings are made up to 500 ml. and returned to the nickel dish. For the colorimetric estimation, 50 ml. of the above solution are treated with 8.4 ml. of 2 *N* nitric acid in a 150-ml. beaker, diluted to about 100 ml., treated with 2 ml. of ammonium molybdate solution, and allowed to stand for 10 minutes. An equal volume of water is titrated in a beaker of the same dimensions with a standard picric acid solution until the colours of the two liquids are the same, and the silicon-content is then read from standard curves. Alternatively, the colour may be measured in a Pulfrich photometer. For the preparation of the standard curves 5 g. of the sodium carbonate and borax mixture is melted in a platinum crucible, which is then heated with water in a nickel dish. The solution is cooled, made up to 500 ml. in a volumetric flask, shaken, and returned to the dish. A solution of 37.2 g. of pure potassium alum per litre is also made up. Twenty-five ml. of the alum solution are added to 25 ml. of the carbonate-borax solution in a 150-ml. beaker, and the solution is treated first with 8.4 ml. of 2 *N* nitric acid and then with 2 ml. of 10 per cent. ammonium molybdate solution, diluted to 100 ml., shaken, and allowed to stand for 10 minutes. Titration of this solution gives the zero point of the curves, and the procedure is repeated with quantities of standard sodium silicate solution corresponding to 0.02, 0.035, 0.05, 0.065, 0.08, 0.10, and 0.11 per cent. of silica. It is important not to grind the sample in an agate mortar, to avoid prolonged contact of the alkaline solution with glass, to use fresh ammonium molybdate solutions (not more than one week old), to have absolutely clear solutions, and to use the correct quantity of nitric acid (50 ml. of the solution containing no silicon is titrated with 2 *N* nitric acid in the presence of methyl red; the volume of acid used plus 4 ml. is the correct volume for 100 ml.). The method has also been adapted to the determination of silica in fluorides.

E. M. P.

Analysis of Chrome Green and Similarly-prepared Pigments. J. H. Van Der Meulen. (*Chem. Weekblad*, 1939, 36, 855-859).—Commercial chrome green is a mixture of chrome yellow and Prussian blue. It sometimes contains lead sulphate also, and in the procedure now described (which is a modification of that suggested by Kappelmeier, *Rec. Trav. Chim. Pays-Bas*, 1931, 50, 711) the presence of this salt is assumed; it is also assumed, for the purposes of calculation, that Prussian blue has the composition $\text{Fe}_4(\text{FeCy}_6)_3$. Eight g. of an intimate mixture of 2.5 g. of the sample and 7.5 g. of pure anhydrous sodium carbonate are treated with 50 ml. of hot water, and the suspension is boiled until a yellow-brown precipitate in a yellow solution is produced. In this way the lead chromate gives lead carbonate and sodium chromate; the lead sulphate gives lead carbonate and sodium sulphate; and the Prussian blue gives sodium ferrocyanide and ferric hydroxide, carbon dioxide being evolved. The mixture is diluted to 100 ml. with hot water and stirred for 15 minutes, and then placed on the water-bath for 10 minutes. Any organic matter is removed by the addition of 10 ml. of a 0.1 *N* solution of potassium permanganate, followed by 15 ml. of 0.1 *N* hydrogen peroxide, which reduces any ferricyanide to ferrocyanide (if the peroxide contains sulphate, 250 mg. of sodium perborate may be used). When the precipitate has settled the solution is filtered, and the residue is washed with hot water. To the combined filtrates are added 5 g. of sodium carbonate and the volume is made up to 500 ml. To 100 ml. of this are added 10 ml. of *N* potassium iodide solution, 25 ml. of 5 *N* hydrochloric acid, and after 1 minute, 5 ml. of a 0.5 *M* solution of zinc sulphate and 100 ml. of water. The iodine liberated is titrated with 0.1 *N* sodium thiosulphate solution with starch as indicator (1 ml. = 10.7743 mg. of lead chromate). The Prussian blue is determined in terms of the sodium ferrocyanide present in the filtrate, the principle of the method being as follows:—In presence of acid part of the chromic acid produced in the filtrate oxidises the ferrocyanide to ferricyanide. If, however, a known quantity (and an excess) of ferrocyanide is added, the chromic acid is reduced quantitatively and the excess of ferrocyanide may be back-titrated with a permanganate solution. Consequently to 100 ml. of the filtrate are added 100 ml. of water, 10 ml. of 10 *N* sulphuric acid, and 25 ml. of a 0.1 *N* solution of potassium ferrocyanide, and the mixture is then titrated with a 0.1 *N* solution of potassium permanganate with erioglaucin-A as indicator. Then the difference between the total volume of oxidising agents present (*i.e.* chromate plus permanganate) and the volume of ferrocyanide added (in ml. of 0.1 *N* solutions) gives the volume of 0.1 *N* ferrocyanide formed from the Prussian blue. Alternatively, 100 ml. of the filtrate are acidified with 10 ml. of 10 *N* sulphuric acid, and a slight excess of 0.1 *N* hydrogen peroxide is added and subsequently removed by means of a 0.1 *N* potassium permanganate solution (with 3 drops of a 0.1 per cent. solution of erioglaucin-A as indicator). The iodine liberated on the addition of 5 to 10 ml. of *N* potassium iodide solution and 5 ml. of a 0.5 *M* solution of zinc sulphate is then titrated with 0.1 *N* sodium thiosulphate solution in the usual way. The lead sulphate is determined by acidifying 100 ml. of the original filtrate with 20 ml. of 5 *N* hydrochloric acid, and adding 5 ml. of *N* (sulphate-free) hydrogen peroxide or 400 mg. of sodium perborate, and 3 ml. of *N* sodium nitrite solution. The sulphate may then be

precipitated from the boiling solution by means of 50 ml. of a 0.05 *M* solution of barium chloride, and determined in the usual way. The total lead-content is determined gravimetrically as lead chromate, 2 g. of the dry mixture of pigment and sodium carbonate being boiled with 25 ml. of water until particles of lead chromate are no longer visible; 10 ml. of 5 *N* potassium hydroxide solution are then added, and boiling is continued until the brown ferric hydroxide is produced, after which 50 ml. of hot water are added. The mixture is warmed and filtered, and the residue is well washed with hot water; it may be retained for a determination of the iron in the usual way. The filtrate is added, with stirring, to a hot mixture of 100 ml. of *N* acetic acid, 10 ml. of *N* potassium dichromate solution and 50 ml. of water, care being taken to avoid loss due to the carbon dioxide evolved. The solution is boiled until the yellow precipitate becomes orange and is then allowed to stand for 2 hours, after which it is filtered, and the residue is washed well with 0.1 *N* acetic acid and then with water and dried at 100° C. until constant in weight.

J. G.

Microchemical

Micro-electrolytic Determinations in Small Volumes. J. Donau. (*Mikrochem.*, 1939, 27, 14–20.)—Two procedures are described, one for very small volumes down to a single drop, the other for larger volumes of the order of a few ml. In the first method the electrolysis is carried out directly in the shallow cup-shaped cathode, made out of platinum foil (0.1 mm. thick), with a handle of 0.5 mm. platinum wire. The dimensions of the electrode are determined by the volume of liquid to be electrolysed. The anode consists of platinum wire coiled horizontally at the tip, the coiled portion being immersed in the liquid. The anode and cathode terminals are fixed to the arms of a U-shaped vulcanite holder. The liquid to be analysed is either measured or weighed into the cathode. In the latter instance the cathode must be covered with a well-fitting lid during weighing. Electrolysis is carried out by passing a current of 2–3 milliamps; at this low current the loss by bubbling is negligible. If necessary, heat may be applied, but drops of water must be added to replace any loss by evaporation. Electrolysis is usually complete in 10 to 20 minutes. The vulcanite holder is rotated through 45° and washing is carried out without interruption of the current. When the ammeter shows no more current passing, washing is complete. The cathode may then be removed, dried on filter-paper and finally on an aluminium block heated to 110° C. After cooling, the cathode is weighed in the usual manner. Both a Kuhlmann balance and the author's modification of the Nernst torsion balance were used (Donau, *Mikrochem.*, 1933, 13, 155; Abst., ANALYST, 1934, 59, 136). If necessary the metal may be dissolved and the electrolysis repeated. When working with larger volumes an apparatus similar to that used by Pregl is preferable. The container is, however, provided with a tap, thus allowing the electrodes to be rinsed without interrupting the passage of the current. Excellent analytical results were obtained by both methods.

J. W. M.

✓ **1.2-Diaminoanthraquinone-3-sulphonic Acid as a Reagent for Copper.** H. E. Ballaban. (*Mikrochem.*, 1939, 27, 57–63.)—The reagent is extremely selective for copper in alkaline solution. When the test is made on the spot-plate,

a drop of the neutral or acid test solution is mixed with a drop of the reagent solution (0.05 g. of 1,2-diaminoanthraquinone-3-sulphonic acid in 100 ml. of water; the solution is stable). When the liquid is made alkaline with sodium hydroxide the colour changes from red-violet to cornflower blue. For very small amounts a blank test is advisable. *Limit of identification*: 0.02% of Cu; *concentration limit*: 1:2,500,000. The test may also be carried out on impregnated paper. In presence of cobalt and nickel, which interfere, the copper salt is first converted into the insoluble thiocyanate on filter-paper. The cobalt and nickel salts are then washed out of the paper, and when the paper is dry the test may be carried out successfully. The method has been applied to the detection of copper in minerals and alloys, and a number of examples are given. Ammonium salts interfere and should be removed prior to the test. J. W. M. ✓

Detection of Ammonia, Calcium and Strontium with Organic Nitro Compounds. R. Fischer. (*Mikrochem.*, 1939, 27, 67-75.)—Organic nitro-compounds may be employed as reagents for ammonia; picric and styphnic acids are especially suitable, the acids being soluble in chloroform, whilst the ammonium salts are insoluble, and may therefore be readily separated from the reagent. Ammonium picrate and ammonium styphnate are identified by means of micro sublimation and micro-determination of melting-points; the former compound sublimes readily at 170-188° C., the latter at 170-190° C. *in vacuo*; the melting-point of the sublimate is determined; 0.1% of ammonia may be detected. Picrolic acid forms very characteristic calcium and strontium salts. The crystals obtained after cautious evaporation on a microscope slide are washed with chloroform and water. The calcium salt melts at 250-270° C., but not sharply; the strontium salt does not melt up to 350° C. As little as 0.2% of calcium or strontium may be detected by this method; four photomicrographs are given. J. W. M.

Micro-determination of Ethyl Alcohol in Pharmaceutical Products: Determination of the Chromic Index. A. Ionesco-Matiu, C. Popesco and O. Constantinesco. (*J. Pharm. Chim.*, 1939, 30, 252-263.)—The authors have made a micro-determination of alcohol in pharmaceutical preparations by a modification of the Nicloux method (*cf. Bull. Soc. Chim. biol.*, 1931, 13, 859). The method is very satisfactory for preparations containing 0.1 to 0.3 per cent. of alcohol; preparations rich in alcohol are diluted, 1000-fold if necessary, before analysis. One ml. of the dilute solution and 1.5 ml. of conc. sulphuric acid are shaken well in a test-tube and titrated on the boiling water-bath with standard potassium dichromate solution at the rate of 1 drop per minute, a micro-burette being used. One drop of methylene blue leuco-base solution, used as external indicator, gives a sharp end-point with a small drop of the solution titrated. The indicator is prepared as follows:—To 1 ml. of a solution of methylene blue (0.05 g. in 25 ml. of glycerin and 75 ml. of water), 3 drops of 10 per cent. sodium thiosulphate solution and 10 drops of 1 per cent. sulphuric acid are added, and the mixture is shaken well and allowed to stand until completely decolorised (about 1 hour). In the dark it can be kept for a few days. One ml. of the standard dichromate solution is equivalent to 1 mg. of alcohol.

Chromic indices of oxidation have been found for (a) the original preparation, (b) the distillate from it, (c) the distillate from acid and alkaline solutions successively. They are known respectively as the total, partial and alcoholic chromic indices of oxidation. The total index is the number of ml. of the standard potassium dichromate solution consumed by 1 mg. of the preparation itself; it is a measure of all the substances oxidisable with chromic acid. The partial index is a measure of the alcohol and volatile material, and the alcoholic index (unless the preparation contains a substance which is retained neither by acid nor alkali on distillation) is a measure of the alcohol only. As each index is a constant for any standard pharmaceutical product, determination of the three constants serves as a test of the quality of a sample. A table of these constants for twenty-eight medicaments is given; determinations of alcohol by the Westphal-Mohr balance, pycnometer, and original Nicloux method are also included. For the analysis, approximately 1 ml. of the product itself is sufficient. E. B. D.

Physical Methods, Apparatus, etc.

Separation of the Components of a Mixture by Fractional Centrifuging.

A. Gourevitch. (*Ann. Chim. anal.*, 1939, 21, 291.)—Separation of a chemical substance from a mixture is often difficult when its chemical properties are imperfectly known. Preliminary experiments were made to determine if separation can be effected by means of a determinable physical property of the body. If, for example, a mixture of salts in solution is evaporated to dryness, the residue, as a rule, will be a mass, more or less homogeneous in appearance, but consisting of crystals, sometimes of microscopic dimensions. If it were found possible to separate the various kinds of crystals occurring in the residue by means of a characteristic physical property (*e.g.* density), a separation of the components of the mass into pure chemical substances would be possible. To investigate the practical application of this principle, it was applied to the residue obtained by the evaporation of an aqueous solution of potassium chloride and potassium dichromate. The residue was placed in a flask with about nine times its weight of thymol and some glass beads, and the flask was shaken in a rotary agitator for three or four days. Microscopical examination of the yellow, compact mass showed it to consist of the débris of the crystals of the two salts suspended in the thymol. If the pieces are sufficiently small, each will consist of one salt or the other. A portion of the ground mass was mixed with a relatively large amount of a mixture of bromoform and nonane, which dissolves the thymol but not the salts. The calculated proportions of bromoform and nonane were such that the density of the liquid after solution of the thymol was 2.20 to 2.40, *i.e.* between that of potassium chloride (1.99) and that of potassium dichromate (2.69). The mixture was then centrifuged at low speed. It was found that the potassium chloride floated on the surface of the liquid and the potassium dichromate formed a sediment at the bottom. A satisfactory separation of the two salts was thus effected, and the practical application of the principle appears to be realisable. A. O. J.

Immersion Liquids for Determination of Refractive Index of Non-opaque Minerals. E. P. Kaiser and W. Parrish. (*Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 560-562.)—The total range covered is n_D 1.411 to 1.785, and the following mixtures are suggested for intermediate ranges:—(1) n_D 1.411 to 1.465: *n*-decane and "medium government oil" mixtures, giving a straight-line curve of n /composition. A similar series in which kerosene fractions were used instead of "medium government oil" has been employed by Butler (*Amer. Mineral.*, 1933, 18, 386). (2) n_D 1.470 to 1.630: "medium government oil"— α -chloronaphthalene, straight-line mixing curves. (3) n_D 1.635 to 1.735: α -chloronaphthalene and methylene iodide; mixing curve is not a straight line and should be determined by experiment (the curve is reproduced in the original paper). (4) n_D 1.740 to 1.785: methylene iodide and solution of sulphur in methylene iodide; straight-line mixing curve. The values of n_D are at 22° C. S. G. C.

Reviews

CHEMICAL SPECTROSCOPY. By WALLACE R. BRODE. Pp. xi + 494. London: Chapman & Hall, Ltd. 1939. Price 36s.

The lack of available material in English has led to the publication of Professor Brode's book, which is apparently based upon the author's experience of courses in chemical spectroscopy at Ohio State University. It gives directions for setting up a laboratory for spectroscopic work and includes a chapter on laboratory experiments in applied spectroscopy. There can be little doubt that if this course of experiments is followed intelligently a considerable knowledge of the practice of spectroscopy for chemical purposes will be obtained. Chapter II gives a very short and condensed account of the theory and nomenclature of atomic and molecular spectra. Without considerable amplification of this chapter it seems unlikely that a beginner could understand and use the outline of the subject presented here. As, however, the reader has his attention called to such works as that of Candler on "Atomic Spectra," it is possible that the chapter may be considered enough for introductory purpose. The chapters dealing with apparatus and methods, and directions for calibration, identification of lines and choice of standard methods are full and adequate, as are those dealing with absorption spectra and the methods of determining them. Chapters VI and VII give an indication of the results of modern theory. For the chemist who desires to use spectroscopic methods in general work this theoretical discussion may be regarded as sufficient. The table of persistent lines, of principal lines in order of wave-length, and of the lines of the elements under different modes of excitation will be found most useful. A valuable set of charts shows the lines of the iron spectrum. A feature of these charts is the inclusion of the principal lines of other elements; this will save a large amount of work in identifying an unknown element. To use these charts to the best advantage it would appear desirable, in practice, to enlarge negatives containing the standard iron lines and the unknown spectrum to the size of the reproductions given in the charts.

The author has realised that the fields of interest of workers may be different, and he indicates at the outset those parts of the book which various workers may

find useful. In the reviewer's opinion, Chapters III to XI should be read by all chemists who are taking up the subject of spectroscopy. So far as can be seen there are few errors, although the "Paschen-Back" effect is here given as "Paschen Bach." The illustrations are in general well done, but some of the reproductions of line spectra are by no means clear, for example, Figure 4-2, 8-15 and 8-50. The index is good and there is an adequate bibliography, most of the chapters having a smaller bibliography on the relevant subject matter. There are "Inserts" containing coloured cellophane sheets for exercise, and two excellent film negatives of samples examined.

It will be remembered that this Society held a discussion on Spectrum Analysis in 1935, and from the wide interest shown on that occasion it was evident that some guide was needed for laboratory use on the method in general. The volume before us may be considered to fill this need for an authoritative statement by a well-known worker in spectroscopy, who knows the difficulties that have to be met and can from his own experience guide the reader. J. J. Fox

A TEXT-BOOK OF QUANTITATIVE CHEMICAL ANALYSIS. By A. C. CUMMING and S. A. KAY. Pp. xv + 496. Seventh Edition. Revised by F. C. GUTHRIE and J. T. NANCE. London: Gurney & Jackson. 1939. Price 15s.

The revision of the new edition of "Cumming and Kay" has consisted, for the most part, in bringing the work into line with modern practice by the introduction of new matter, for which space has been provided by deleting the section on organic analysis; the new edition contains fourteen pages more than the sixth. Methods for the determination of lithium, molybdenum, platinum, tungsten and vanadium have been added. Among the more recently introduced methods of procedure now included may be mentioned the use of diphenylamine as internal indicator in the dichromate titration of iron, of ceric sulphate as a volumetric standard solution, and of sodium rhodizonate as indicator in the volumetric determination of sulphate, the determination of fluoride with thorium nitrate, the determination of tungsten by precipitation with benzidine, the determination of titanium with tannin and phenazone and of hardness in water with potassium palmitate. The section on electrometric methods has been extended to include oxidation-reduction systems.

Two useful determinations that were mentioned as missing by the reviewer of the sixth edition (ANALYST, 1935, 60, 503) have now been included—the colorimetric determination of bismuth and the direct determination of sodium by means of zinc uranyl acetate. In the former method no provision has been made for the usual precaution against possible interference by free iodine.

The work of revision has been carried out with thoroughness, leaving but little room for criticism of the present text. There is an ambiguity on page 344, where the statement that "tungstic acid can also be precipitated from alkali tungstate solutions obtained after fusion with alkali hydroxide or carbonate" might be read as meaning that the precipitation is sufficiently complete to form the basis of a method for determination; no such method is, of course, described. The method given for the reduction of stannic chloride by means of metallic antimony, as a preliminary to titration by iodine, has been abandoned by most routine workers as unreliable, probably on account of after-reduction or to action of iodine on the

remaining antimony powder. Among metallic reducing agents for tin, the most favoured at the present time would appear to be aluminium, with nickel, iron and lead for special purposes. The description of the colorimetric determination of titanium contains no mention of suppression by means of phosphoric acid of the colour due to ferric chloride—a modification that sometimes has its uses. The bibliography in the appendix appears to have been overlooked in revision. Thus the authors appear to be unaware that *THE ANALYST* changed its publishers in 1922.

The book is intended for use at the bench, and therefore contains no general theoretical matter. It is written in an easy style and is practically free from typographical error. The pagination, type and binding constitute an excellent example of the art of book-making.

F. L. OKELL

THE BRITISH ENCYCLOPAEDIA OF MEDICAL PRACTICE. Vol. 12. Butterworth & Co. (Publishers), Ltd. 1939. Price 35s.

- (1) TOXICOLOGY. I. HOMICIDAL, SUICIDAL AND ACCIDENTAL POISONING, by G. ROCHE LYNCH and D. M. PRYCE. II. INDUSTRIAL POISONING, by DONALD HUNTER.
- (2) URINE EXAMINATION, by J. DOUGLAS ROBERTSON.
- (3) VITAMINS, by LESLIE J. HARRIS.

An encyclopaedia of medical practice is compiled, primarily, for the general practitioner and not for the expert in any particular branch. It should provide a ready means of finding such known information as may be necessary for the proper understanding and treatment of any medical problem arising in daily routine. The authors must find it difficult to decide how much to include or, possibly with some subjects, how much to omit. The three articles under review have been written by recognised experts and every confidence may be placed in their selections.

(1) These two monographs deal with two different and distinct branches of Toxicology. One is of rare immediate interest to most practitioners, but when a case arises it is usually a matter of life or death; the other is the concern of many.

Drs. Roche Lynch and Pryce begin with four pages of introduction, being general advice to the physician. The importance of samples, either for diagnoses during life or to establish the cause of death, is stressed, and general instructions are given for taking them. It was to be expected that the experience of the authors would place this matter first. Points to be borne in mind during diagnosis are next enumerated; the account is short so that it can be quickly read and to the point so that it can be easily remembered. The remaining 60 pages deal with about 40 substances or groups of substances classified as follows:—

- (1) Gases (carbon monoxide and a cross reference to war gases).
- (2) Corrosive acids and alkalis.
- (3) Synthetic organic substances (other than alkaloids).
- (4) Alkaloids.
- (5) Cantharides.
- (6) Inorganic and metallic poisons.
- (7) Fungi.
- (8) Abortifacients.
- (9) Powdered glass.

The general plan is to treat under each poison the physical properties, fatal dose, morbid anatomy, symptoms and treatment. All the commoner poisons which have caused death in recent years are included, except iodine, chloroform and cinchophen. The omission is probably intentional, since iodine and chloroform are readily recognised, and cinchophen, being now a Fourth Schedule poison, can

be obtained only on medical prescription. In addition these poisons are dealt with specifically in other monographs, such as those covering anaesthetics, toxic jaundice, etc.

The language is clear, and the general use of short, pithy sentences devoid of superfluities and jargon seems to accentuate the facts. Under carbon monoxide there is a liability to confusion in certain instances as to whether percentage refers to blood saturation or to the gas breathed. It would be interesting to know the indicator, a sort of universal "toxicator," suggested by the sentence, "If it is thought that the medicine is being tampered with, the prescription may be modified so that the addition of poison will be betrayed by a change of taste or of colour."

Dr. Hunter opens with notes on the prevention of disease in industry and on legal notification and compensation. This gives much valuable information, particularly useful for the practitioner who is confronted but rarely with industrial illnesses. Then follow particular sub-monographs on arsenic, mercury, silver, manganese, toxic gases, benzene and homologues, nitro and amino derivatives, chlorinated hydrocarbons, other organic compounds (carbon disulphide, acetone and dioxan), injuries from X-rays and injuries from radio-active substances.

It would appear that in each instance there is a good and concise account both as regards the clinical picture and preventive treatment.

Industrial poisoning may be regarded as an important but minor part of industrial diseases and the present article must be considered in conjunction with those on Occupational Diseases, certain Skin Diseases and Lead Poisoning appearing elsewhere in the encyclopaedia.

(2) Dr. Robertson's article gives details of the tests which may be applied in the routine examination of urine. These comprise colour (pigments and drugs), quantity, specific gravity, reaction, albumin and blood, casts, chemical constituents (oxalates, chlorides, sulphates, urea, uric acid, bile, ketone bodies, alcohol and sugar) and tests for renal function.

(3) Dr. Harris first sketches the general historical background relating to vitamins and classifies them broadly on their solubility. He then deals with the more important ones individually or as complexes, discussing history, chemical nature and properties, distribution in food, methods of assay, physiological action, incidence and clinical picture of deficiency diseases, diagnosis, prophylaxis and curative treatment, dosage, daily requirements and special needs. At the end are short notes on dietetics and applications. In the short space of 29 pages a comprehensive survey of vitamins has been attempted and has been achieved.

J. R. NICHOLLS

THE SOCIAL FUNCTION OF SCIENCE. By J. D. BERNAL, F.R.S. Pp. xvi + 482, with 3 charts. London: Routledge. 1939. Price 12s. 6d. net.

In this book Professor Bernal has set out the results of his attempts to discover to what extent science is practised in this country, to what extent that practice is organised and by whom, and to what extent the results of its practice, whether co-ordinated or not, find expression in the machinery of our social and daily life. This has involved him in a survey of a kind never before undertaken, to the best of my knowledge; at any rate, the results of such a survey have never before been published.

After some general observations (Part I: What Science Does) of an introductory and historical nature, the author proceeds to examine the existing organisation of scientific research in this country and then the part played by science in education. Next he discusses the efficiency of scientific research and the application of science, with a separate chapter on its application to war purposes. This part of the book concludes with a chapter on International Science.

The second part of the book (Part II: What Science Can Do) is of a more controversial nature. It ranges over the whole of the matters discussed in Part I and more beside. It discusses such details as revising the scientific curriculum in schools, colleges and universities; the function of scientific publications and the facilitating of travel for scientists; science in relation to the economic organisation of industry and finance; science in relation to individual human needs, such as food, clothing, housing and so on; science in relation to industries producing goods or services; and science and culture.

No review can do justice to the scope and detail of Professor Bernal's book and to the amount of investigation that its preparation must have involved. Indeed, it has involved more than that, for the information, collected from a widely scattered mass of heterogeneous sources, has needed the integrative powers of a singularly acute mind—coloured maybe by certain clearly envisaged political principles or preconceptions!—to work the whole of the material into a picture that can be apprehended, if not comprehended.

Chemists will perhaps be a little surprised at Professor Bernal's remarks (page 43) about the National Chemical Laboratory to which he attributes the function of "assisting the Board of Trade in standardising products from the chemical point of view." From the fact that there is no mention anywhere in the book of the Government Laboratory, it seems that he has here, for once, made a slip over his facts and confused the Teddington institution, important but small, with its much larger and more utilitarian elder brother at Clement's Inn. Moreover, the large amount of research carried out at the older laboratory also must be included in any comprehensive survey of official, or officially organised, science in this country.

This curious confusion is the only error of fact that I have been able to discover; this does not mean that there are not others, and it is to be hoped that readers who discover them will inform Professor Bernal, who will be the first to recognise that the value of his work must be enhanced by every further step taken towards correct presentation of the facts.

When allowance is made for any small errors of fact or minor errors in logic, should there be any, the reader of this book will find himself faced then with some major problems, whose nature cannot possibly be affected by the accuracy or inaccuracy of a statistically insignificant number of minor facts. Is the picture that Professor Bernal draws of organisation, or its lack, in British science, a correct one? Are the alterations that he advocates desirable from the point of view of both science and society? Are the steps he proposes for such re-organisation themselves practicable within the present social structure? If not, are the kind of alterations to that structure envisaged, and sometimes depicted, by Professor Bernal possible and desirable methods of obtaining the object in view? Some of

those who are politically minded will agree with Professor Bernal in all his answers to these questions. Some will agree with some of his answers and not with others. Some may agree with none of his answers and on them will be the onus of supplying alternatives. But to those who hold the view, in the teeth of Professor Bernal's evidence, that the interests of science and scientists can be considered apart from politics in general, and from party politics in particular, there will still be open a course that should commend itself, and that is to use this book as a source of facts and information, marshalled with great industry and skill, and to take its author's opinions as red!

A. L. BACHARACH

THE MINERAL WATER TRADE YEAR BOOK. Published by the National Union of Mineral Water Manufacturers, Ltd., Great Charles Street, Birmingham. Pp. 194. 1939.

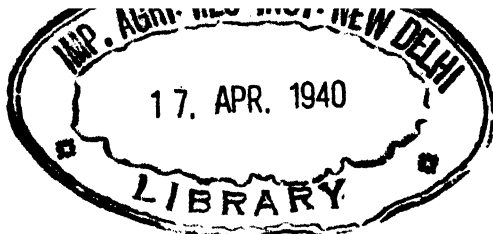
Early in the present century the mineral water industry received an unpleasant shock when it was found that a large proportion of the soda water sold in London was bacteriologically very impure. This discovery led to far-reaching hygienic changes in mineral water factories and had much to do with putting an end to the rule-of-thumb methods that had for so long been in use. To-day the industry, following the lead of the brewers and food manufacturers, has organised itself into an association which is concerned not solely with material benefits for its members, but also with supplying products of unquestionable purity to the public. For example, a scheme for taking steps towards formulating standards for mineral waters has recently been sanctioned by the National Union.

This is one of the questions discussed in the first edition of the *Trade Year Book*, which appears to have omitted nothing of essential importance to the industry and is thoroughly practical in its outlook. Factory law is dealt with in a useful section which summarises the changes brought about by the Factories Act, 1937. Then come sections on Workmen's Compensation, Electricity Regulations, Road Traffic, Shop Legislation, Table Water Duties, Trade Marks and the Merchandise Marks Act. On the more technical side there are sections on Bottle and Stopper Cleaning, Carbonation and Bottling, Recipes for the Syrup Room, Water Purification and the Use of Saccharin. Lastly, there are miscellaneous sections on such subjects as Cost of Living, National Insurance, First Aid Hints and so forth.

Among the special articles is one contributed by a Scottish lawyer, Mr. T. Young, on the liability of the manufacturer to the consumer. This is a legal discussion on the bearing of the case of *Donaghue v. Stevenson*, which arose out of a snail that had introduced itself into a bottle of ginger beer, and finally, after the case had reached the House of Lords, determined the question of the liability of a manufacturer to a member of the public with whom he had had no contact.

Another article of practical value is by an engineer, Mr. Scott Hall, who discusses the factors for the efficient management of motor transport. From this brief outline of its contents it will be seen that this Year Book will become a *vade mecum* for all mineral water manufacturerers, while the chemists whom they employ or consult will also be able to turn to it for help in some of their difficulties.

EDITOR



THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

A JOINT MEETING with the Food Group of the Society of Chemical Industry was held at the rooms of the Chemical Society, Burlington House, W.1, on February 7th.

In the absence of the President, Professor W. H. Roberts, the chair was taken by Dr. Roche Lynch, and the Chairman of the Food Group, Mr. E. B. Anderson, was also on the platform.

The subject was "Carotene and Allied Substances in Foods and Feeding Stuffs," and the following papers were read and discussed:—"The Constitution and Physiological Significance of Carotene and Allied Pigments," by R. A. Morton, D.Sc., F.I.C.; "The Commercial Determination of Carotene and Allied Pigments, with Special Reference to Dried Grass and other Leafy Materials," by W. M. Seaber, B.Sc., F.I.C.

The following candidates have been elected members of the Society:

Horace George Battye, Ph.D., F.C.I.C., Registered Engineer (Chemical), Ontario.

George Malcolm Dyson, B.A., B.Sc. (Oxon), Ph.D. (Lond.), F.I.C., A.M.I.Chem.E., Chief Chemist, Genatosan, Ltd., Loughborough, Leicestershire. (*Through North of England Section.*)

Jack Kerfoot, B.Sc., A.I.C., Assistant Chemist, Research Department, L.M.S. Railway Co. (*Through North of England Section.*)

Ada Frances McColl (Miss), A.I.C., Chemist in department of Scottish Co-operative Wholesale Society. (*Through Scottish Section.*)

NORTH OF ENGLAND SECTION

THE Fifteenth Annual General Meeting of the Section was held at Manchester on January 27th, 1940. The Chairman (Professor T. P. Hilditch) presided over an attendance of thirty-two. The President, Professor W. H. Roberts, was present.

At the request of the Chairman the members stood in silence as a mark of respect to the late H. T. Lea.

The Secretary presented the Report and Financial Statement, which were adopted.

The following appointments were made:—*Chairman*, J. R. Stubbs; *Vice-Chairman*, W. G. Carey; *Committee*, H. Childs, H. H. Jones, T. W. Lovett, H. M. Mason, J. G. Sherratt, C. J. H. Stock; *Honorary Auditors*, U. A. Coates, J. W. H. Johnson; *Honorary Secretary and Treasurer*, A. Lees.

The following papers were read and discussed:—"Some Aspects of the Purification of Polluted Waters for Industrial Use," by J. G. Sherratt, B.Sc., F.I.C., and "The Estimation of Lead in Drinking Waters," by C. H. Manley, M.A., F.I.C.

Estimation of Praseodymium and Neodymium in Solution from their Absorption Spectra

BY J. NEWTON FRIEND, D.Sc., AND DOUGLAS A. HALL, Ph.D.

THE absorption spectra of solutions of the rare earths, particularly praseodymium and neodymium, have been studied by many investigators, both in the visible region, when possible, and photographically in the ultra-violet region. One of the most important objects has been the detection and estimation of small quantities of an earth present as impurity in excess of some other earth. This is of special value in atomic weight determinations when earths of exceptional purity are required, while purely chemical methods of separation are either not available or give uncertain results.

The method breaks down when the bands of one element overlap those of another. Thus a neodymium band readily obliterates the most characteristic erbium band, $\lambda = c.5230$, so that the latter earth may be overlooked in presence of only small amounts of neodymium (Driggs and Hopkins¹). Although a certain amount of overlapping occurs with praseodymium and neodymium, the difficulty may be partly overcome by a suitable selection of the bands to be studied.

In general, three different methods of estimating the rare earths by their absorption spectra in solution have been attempted. Delaunay² examined solutions of the nitrates of praseodymium and neodymium, plotting curves to show the relation between the widths of the chief bands in the visible region and the lengths of the absorption tube. These widths were compared with those given by solutions of unknown composition and, on the assumption that Beer's law is obeyed, the concentrations of the salts giving bands of equal widths were taken as inversely proportional to the length of the absorption tube. Panchromatic plates were used, and an accuracy of some 5 per cent. was claimed. A weakness of this as a general method lies in the diffuseness of many bands, especially in dilute solution.

A second method, suggested by Yntema,³ surmounts this difficulty. It consists in finding the dilution necessary to effect the disappearance of the most persistent band of a given earth in a mixture under rigid photographic conditions and calculating the original concentration from that of the pure rare earth solution which likewise just fails to give the same band under identical conditions. Inoue⁴ found that traces of cerium in the presence of a large excess of other rare earths can be estimated with considerable accuracy in this way, using the band with

maximum absorption at 2469 Å. We tried this method in the visible region with solutions of neodymium, using the unaided eye instead of photographs. It was found, however, that the eye tired, so that the results were uncertain.

The third and oldest method is based on the Nessler principle and, when confined to the visible spectrum, does not require a camera. It consists in diluting the unknown solution until the intensities of suitable absorption bands appear, by direct vision, to equal those of a standard solution placed in juxtaposition. Both Brauner⁵ and Jones⁶ used this method in estimating small quantities (up to some 3 per cent.) of praseodymium as impurity in neodymium solutions, and *vice-versa*. This procedure has been widely adopted in estimating traces of these and other coloured earths in modern atomic weight determinations.

All of these methods are based on the assumption that the spectrum is not appreciably affected by the presence of other earths or compounds in the same solution. Inoue⁴ stated that this was true of the ultra-violet bands of cerium in presence of excess of other rare earths. But it has long been known that the visible bands of praseodymium and neodymium are materially affected by the nature of the acid radicle present in both position and intensity. Further, the bands of the nitrates are influenced by the presence of other rare earth nitrates, by magnesium nitrate and even by excess of free nitric acid (Quill, Selwood and Hopkins⁷; Selwood⁸). It is true that Inoue⁴ denied this in respect of chloride solutions, but no details of his apparatus are given and no data concerning the concentrations and proportions of the added salts. It is impossible, therefore, to gauge the sensitiveness of his procedure. Schaeffer⁹ and, more recently, Uzumasa¹⁰ have observed that organic solvents also affect the bands. These results support the destructive criticisms of Baxter and Chapin¹¹ on the supposed accuracy of the estimation of praseodymium by the third method given above.

When mere traces of impurity are concerned, such as the 0.005 per cent. of holmium in the yttria used in an atomic weight determination (Kremers and Hopkins¹²), an error of even 20 per cent. might still be without influence on the result. But with higher concentrations, such as those used by Brauner and by Jones, mentioned above, a similar percentage error might be serious. No determination of the magnitude of this error appears to have been made. The present research has been undertaken to ascertain the order of accuracy attainable by the third method, using pure solutions of the earths, and also the toleration concentrations of free nitric acid and the nitrates of cerium and magnesium that may be present without appreciably affecting the results. The present study is limited to the nitrates of praseodymium and neodymium, and the results have proved encouraging. The experiments were carried out with a Hilger constant deviation spectrometer illuminated by 40-watt gas-filled opal lamps. The absorption cells were 6 cm. in length and the slit was set at the minimum width to yield a clear spectrum with least eye strain (1.0 on the scale of the Hilger instrument). The concentrations of the rare earth solutions were determined by precipitation as oxalate and ignition to oxide, using the precautions already detailed for neodymium (Friend¹³) and praseodymium (Friend¹⁴).

NEODYMIUM NITRATE.—It was first necessary to ascertain which of the numerous absorption bands could be most conveniently studied. Three bands

were eventually selected, namely, band Nd1, extending in 3.5 per cent. solution from 5194 to 5240Å; this, on dilution, yielded eventually two lines* at approximately 5216 and 5203Å. The two other bands yielded lines at approximately 5120 and 5085Å, respectively; these are denoted as Nd2 and Nd3. A further band, Nd4, occurred round 5770Å.

Since, with increasing density of an absorption band, small variations in that density become less easy to detect, solutions of various concentrations were tested to determine the conditions of optimum sensitivity. A standard solution was placed in one cell and band Nd1 was matched spectroscopically with the corresponding band of an unknown solution in a second cell of identical dimensions so placed that the two spectra lay in juxtaposition, the one immediately above the other. The matching was carried out in two ways, namely, "from above," that is, by diluting a more concentrated unknown solution until it matched the standard, and "from below," that is, by increasing the concentration of a more dilute solution. The results obtained were as follows:

TABLE I

Nd(NO₃)₃ g. per litre

| | (1) | (2) | (3) |
|--------------------|------|------|------|
| Standard | 18.3 | 34.7 | 83.2 |
| Matched: | | | |
| (i) From above.. | 18.8 | 35.1 | 81.8 |
| (ii) From below .. | 18.6 | 34.7 | 89.7 |

From the table it is evident that a suitable concentration is afforded by 30 to 40 g. of neodymium nitrate per litre (0.27 to 0.36 *N*) the error being only about 1 per cent. These concentrations have therefore been used throughout this research.

Nitric Acid.—The effect of this acid was determined by taking 25 ml. of standard neutral neodymium nitrate solution in two similar cells, adding distilled water to one and an equal volume of diluted nitric acid to the other. The densities of the absorption bands were compared after each addition. It was ascertained by analysis that, with the concentrations used, there were no appreciable volume changes on mixing.

With unit free acid normality no difference could be detected in the bands; at 1.16 *N* bands Nd1 and 2 began to fade. At higher concentrations they changed not only in intensity but also in structure.

In order to determine the order of accuracy attainable by matching neutral and 2.15 *N* acid solutions, bands Nd2 and 3 were compared because Nd1 became too diffuse. Equal band densities were given with:

3.04 per cent. of Nd(NO₃)₃ in 2.15 *N* nitric acid.

2.36 " " " " neutral solution.

The error was thus 28.8 per cent.

Magnesium Nitrate.—Similar experiments showed that up to a concentration of 0.95 *N* (70.5 g. of Mg(NO₃)₂ per litre) there was no perceptible difference in bands Nd1, 2 or 3. With 1.15 *N* nitrate, band Nd1 was slightly blurred and

* The term refers to the wave-lengths of the imaginary lines that represent the centres of the bands (at great dilution).

weakened. This would affect the estimation. Experiments up to 2 *N* showed that, whilst free nitric acid affected both the intensities and positions of the bands, with magnesium nitrate the bands remained in the same positions, but became paler and blurred.

Sodium Nitrate.—Up to normal concentration, *i.e.* 85 g. per litre, this salt exerted no perceptible influence. A slight effect was observed with 1.03 *N*, and with 2 *N* solutions the differences were pronounced.

Cerous Nitrate exerted no perceptible influence up to 0.6 *N* (65.3 g. of $\text{Ce}(\text{NO}_3)_3$ per litre), but at higher concentrations disturbing effects became pronounced, band Nd1 losing its double structure.

Lanthanum Nitrate.—Band Nd1 retained its double structure up to 1.0 *N* (108.3 g. of $\text{La}(\text{NO}_3)_3$ per litre), although the intensity of the band was sufficiently weakened at 0.8 *N* to affect the estimation of the neodymium; at 0.7 *N* (75.8 g. of $\text{La}(\text{NO}_3)_3$ per litre) the effect was negligible.

The effect of nitrates on the Nd4 band should be recorded. It gained in intensity on addition of nitrates other than neodymium nitrate, whereas Nd1 lost. With 1.0 *N* added nitrate Nd4 was generally darker and 25Å wider than in the pure solution, in which, however, it showed more signs of structure.

PRASEODYMIUM NITRATE.—Similar series of experiments were carried out with solutions of praseodymium nitrate. The most suitable band for study was the one extending from 5980 to 5825Å in 3.86 per cent. solution, yielding on dilution a most persistent line at 5890Å.

As with neodymium nitrate, a suitable concentration was 30 to 40 g. of praseodymium nitrate per litre, with which the matching error was less than 1 per cent. This is shown in Table II.

TABLE II

| | | Pr(NO ₃) ₃ g. per litre | | |
|----------|--------------|--|------|------|
| | | (1) | (2) | (3) |
| Standard | | 38.6 | 24.5 | 93.1 |
| Matched: | | | | |
| (i) | From above.. | 38.6 | 24.3 | 87.6 |
| (ii) | „ below .. | 38.8 | 24.5 | 89.0 |

Nitric Acid.—Addition of nitric acid up to 0.8 *N* (50.4 g. of HNO_3 per litre) had no detectable influence. With 0.9 *N* acid the band weakened appreciably in intensity. Equal band intensities were given with

3.37 per cent. of $\text{Pr}(\text{NO}_3)_3$ in 2.15 *N* nitric acid.

2.80 „ „ „ neutral solution.

The error was thus 20 per cent.

Magnesium Nitrate could be added up to a concentration of 0.8 *N* without affecting the results. In 1.0 *N* solution the band was distinctly less dense.

Cerous Nitrate.—The toleration limit of cerous nitrate was approximately 0.6 *N*. With 0.7 *N* the band was paler and somewhat blurred.

MIXTURES OF THE NITRATES OF PRASEODYMIUM AND NEODYMIUM.—As praseodymium and neodymium invariably occur together in nature, and cannot be quantitatively separated, it is particularly useful to be able to estimate their

amounts in a mixture. Chemical methods, based on the higher oxygen-content of the black oxide of praseodymium, do not yield reliable results (see Prandtl and Ducrue,¹⁵ and Prandtl and Huttner¹⁶). Unfortunately, there is considerable overlapping of the absorption bands. Whilst it is apparently impossible to determine directly with any approach to accuracy the amount of praseodymium nitrate in the presence of neodymium nitrate, the latter can readily be estimated in presence of up to 50 per cent. of the former in 4 per cent. solutions, using bands Nd1, 2 and 3. The following results (Table III) were obtained:

TABLE III

| | | Concentration in g. per litre | |
|---|----|-------------------------------|------|
| Pr(NO ₃) ₃ taken | .. | 13.0 | 19.3 |
| Nd(NO ₃) ₃ .. | .. | 23.5 | 17.6 |
| found | .. | 23.6 | 17.8 |

The error is of the order of 1 per cent. or less.

SUMMARY.—Using a 6-cm. absorption cell, neodymium and praseodymium nitrates can be estimated separately in solutions of 3 to 4 per cent. concentration with an accuracy of about 1 per cent. by comparing the intensities of suitable absorption bands with those of standard solutions.

The toleration limits of other substances that may simultaneously be present in solution without affecting the results are given in the following table (Table IV), in which, for simplicity's sake, the concentrations are stated to the nearest whole gram.

TABLE IV

| | | Nd(NO ₃) ₃ | | Pr(NO ₃) ₃ | |
|-----------------------------------|----|-----------------------------------|-----------|-----------------------------------|-----------|
| | | g. per litre | Normality | g. per litre | Normality |
| HNO ₃ | .. | 63 | 1.0 | 50 | 0.8 |
| Mg(NO ₃) ₂ | .. | 70 | 0.95 | 60 | 0.8 |
| NaNO ₃ | .. | 85 | 1.0 | — | — |
| Ce(NO ₃) ₃ | .. | 65 | 0.6 | 65 | 0.6 |
| La(NO ₃) ₃ | .. | 76 | 0.7 | — | — |

In presence of neodymium, praseodymium cannot be estimated in this way; but the former can be readily estimated in presence of up to 50 per cent. of the latter.

REFERENCES

1. F. H. Driggs and B. S. Hopkins, *J. Amer. Chem. Soc.*, 1925, **47**, 363.
2. E. Delaunay, *Compt. rend.*, 1927, **185**, 354.
3. L. F. Yntema, *J. Amer. Chem. Soc.*, 1923, **45**, 907.
4. T. Inoue, *Bull. Chem. Soc. Japan*, 1926, **1**, 9.
5. B. Brauner, *Chem. News*, 1898, **77**, 161.
6. H. C. Jones, *Amer. Chem. J.*, 1898, **20**, 345.
7. L. L. Quill, P. W. Selwood and B. S. Hopkins, *J. Amer. Chem. Soc.*, 1928, **50**, 2929.
8. P. W. Selwood, *id.*, 1930, **52**, 3112, 4308.
9. H. Schaeffer, *Physik. Z.*, 1906, **7**, 822.
10. Y. Uzumasa, *Bull. Chem. Soc. Japan*, 1931, **6**, 147; 1932, **7**, 85.
11. G. P. Baxter and H. C. Chapin, *J. Amer. Chem. Soc.*, 1911, **33**, 1.
12. H. C. Kremers and B. S. Hopkins, *id.*, 1919, **41**, 718.
13. J. Newton Friend, *J. Chem. Soc.*, 1930, 1633.
14. — *id.*, 1932, 2410.
15. W. Prandtl and H. Ducrue, *Z. anor. Chem.*, 1926, **150**, 105.
16. W. Prandtl and K. Huttner, *id.*, 1925, **149**, 235.

THE TECHNICAL COLLEGE
BIRMINGHAM

December 5th, 1939

The Potentiometric Estimation of Glucose with Potassium Ferricyanide in Sodium Carbonate Solution

By H. T. S. BRITTON, D.Sc., F.I.C., AND LESLIE PHILLIPS, M.Sc., A.I.C.

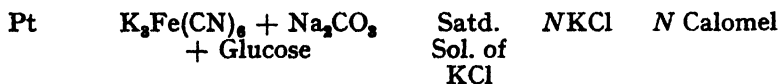
THE fact that potassium ferricyanide oxidises glucose in alkaline solution was first observed by Gentile,¹ but it was not until 1923 that Hagedorn and Jensen² applied the reaction to the quantitative estimation of glucose. They added the glucose to a solution containing an excess of alkaline ferricyanide and heated on a boiling water-bath for 15 minutes. The excess of potassium ferricyanide was then determined by titration by the potassium iodide and sodium thiosulphate method. Van Slyke and Hawkins³ estimated the excess of ferricyanide by measuring the volume of nitrogen evolved as the result of the reaction with hydrazine.

Whitmoyer⁴ adopted a similar method with the exception that, instead of titrating the excess of ferricyanide, he titrated the ferrocyanide formed on oxidising the glucose in the presence of sodium carbonate. This was done by acidifying and titrating with ceric sulphate, with alphazurine G as an internal "redox" indicator.

The direct potentiometric estimation of glucose has hitherto escaped attention, although Wood⁵ has devised a somewhat indirect method based on the extent of the "poising action" (*i.e.* buffer action in regard to the oxidation-reduction process) produced by the equilibrium between ferricyanide-ferrocyanide ions which is set up when a glucose solution is added to an alkaline solution of known concentration of potassium ferricyanide at 100° C.

The work to be described consisted in following the changes in oxidation-reduction potential as glucose is progressively added to an alkaline solution of potassium ferricyanide. Preliminary work appeared to show that sodium carbonate provided the best alkaline medium for the purpose. The titrations were carried out in a boiling water-bath, thereby providing a temperature of 92 to 94° C. in the titration vessel. It was also found that there was no need for the rigid exclusion of air.

EXPERIMENTAL.—The apparatus employed was that already described (Britton and Phillips⁶), and the cell combination was

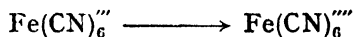


For the titrations, mixtures of *N* sodium carbonate and 0.1 *N* potassium ferricyanide solutions were used. The volumes of each used (given in brackets); their respective concentrations, together with the concentrations of glucose used as titrant, are given in Table I. The original potassium ferricyanide solution, from which the solutions above were prepared, was analysed potentiometrically with potassium permanganate in sulphuric acid solution.

TABLE I

| No. | Concentration of alkaline ferricyanide solution g.-mols. per litre | | Glucose, g. per litre | Amount of glucose solution required ml. | Mols. of $K_3Fe(CN)_6$ per 1 mol. of glucose |
|-----|---|------------|--------------------------|---|---|
| | $K_3Fe(CN)_6$ | Na_2CO_3 | | | |
| 1 | 0.0095 (20) | 0.36 (50) | 1 | 20.33 | 5.89 |
| 2 | 0.0095 (20) | 0.36 (50) | 2 | 10.13 | 5.87 |
| 3 | 0.0095 (20) | 0.36 (50) | 3 | 6.77 | 5.89 |
| 4 | 0.0095 (20) | 0.36 (50) | 4 | 5.07 | 5.90 |
| 5 | 0.0003 (5) | 0.46 (50) | 2 | 2.57 | 5.89 |
| 6 | 0.0055 (10) | 0.42 (50) | 2 | 5.12 | 5.89 |
| 7 | 0.0077 (15) | 0.38 (50) | 2 | 7.72 | 5.88 |
| 8 | 0.0110 (25) | 0.33 (50) | 2 | 12.83 | 5.91 |
| 9 | 0.0125 (30) | 0.31 (50) | 2 | 15.38 | 5.92 |

Figure 1 illustrates some of the titration curves; the numbers by which they are marked refer to the titrations recorded in Table I. As shown by the curves, the potentials set up at the platinum electrode during the initial stage of the titrations were positive with respect to the normal calomel electrode, but as the titrations progressed the platinum electrode became negative. The potentials prevailing during the first stage are governed by the reduction process:



thus at 90–94° C.

$$E_{Pt} = e_{Fe(CN)_6''' \longrightarrow Fe(CN)_6''''} - 0.073 \log \frac{[Fe(CN)_6''''']}{[Fe(CN)_6''']}$$

which shows that as the concentration of ferricyanide-ions diminishes the potential of the platinum electrode becomes increasingly negative. The advantage of potassium ferricyanide over complex alkaline copper tartrate solutions lies in the fact that the potential range corresponding to this process is not so negative as that for $Cu^{++} \rightarrow Cu^+$.

The completion of the reduction of the potassium ferricyanide present in the various solutions is, as Fig. 1 shows, marked by excellent inflexions extending over 400 millivolts for the more concentrated glucose solutions and over 500 millivolts for the more dilute solutions. This sudden drop in potential usually occurs with the addition of 0.1 ml. of glucose solution. The time required to perform these titrations as far as the end-point inflexion is 20 to 30 minutes. The potentials are readily established (within 1 to 3 minutes), and are reproducible. After the end-point, however, the potentials are more sluggish in their attainment and sometimes require as long as 10 minutes before they become constant. This, however, is immaterial to the analytical value of the potentiometric method.

The last column of Table I shows that in the range of concentrations of the solutions used in these titrations the number of molecules of potassium ferricyanide required to oxidise a single molecule of glucose is sensibly constant at 5.9. When compared with complex copper tartrate solutions, in which approximately 5 mols. of CuO are required, it will be seen that alkaline ferricyanide is a little more effective in its action on glucose, in that it causes the oxidation of glucose to proceed to a

greater extent. It is possible that this is responsible for the fact that the potentials set up after passing the end-point are more negative than with Fehling's solution. These more negative potentials, coupled with the higher potentials before the

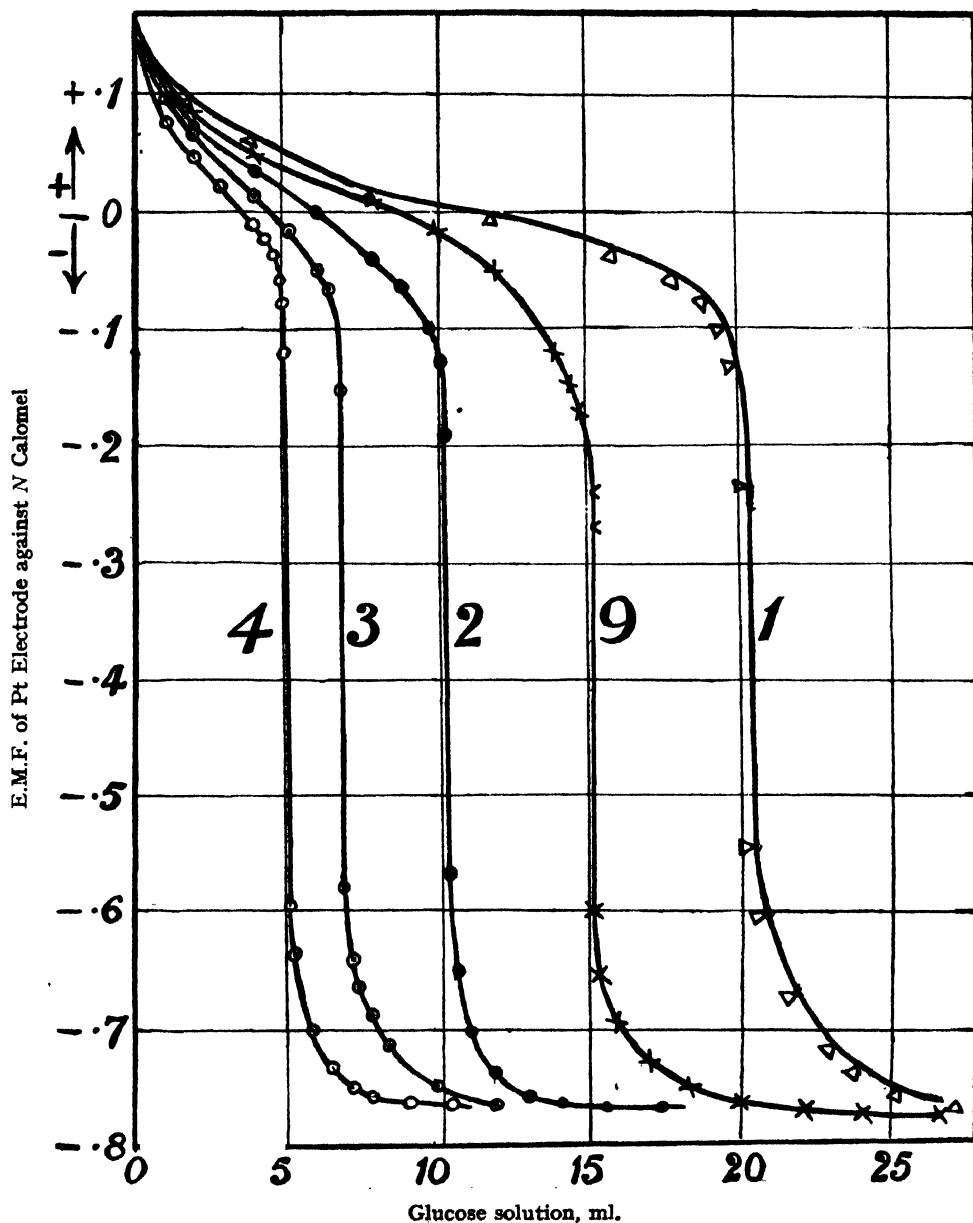


Fig. 1

oxidation of the glucose is complete, give rise to the excellent inflexions obtained and therefore to the greater accuracy of the method, as compared with the Fehling method.

Another point that is brought out by the potentials set up at the end-points and the magnitude of the vertical inflexions is that they pass through the range in which methylene blue becomes decolorised. Methylene blue has been found to be a serviceable internal indicator for titrating sodium carbonate and potassium ferricyanide solutions with glucose at 90 to 100° C. This is being further investigated and will be the subject of a later paper.

One of us (L. P.) wishes to place on record his thanks for the award made to him by the Senate of this College from the Andrew Simons Research Fund.

REFERENCES

1. J. G. Gentile, *Chem. Centr.*, 1859, **30**, 504.
2. H. C. Hagedorn and B. N. Jensen, *Biochem. Z.*, 1923, **135**, 145.
3. D. D. Van Slyke and J. A. Hawkins, *J. Biol. Chem.*, 1928, **79**, 739.
4. R. B. Whitmoyer, *Ind. Eng. Chem.*, 1934, **6A**, 208.
5. W. B. Wood, *J. Biol. Chem.*, 1935, **110**, 219.
6. H. T. S. Britton and L. Phillips, *ANALYST*, 1940, **65**, 18.

WASHINGTON SINGER LABORATORIES
UNIVERSITY COLLEGE
EXETER

July, 1939

A New Test for the Detection of Molybdenum and Tungsten

BY J. HUBERT HAMENCE, PH.D., M.Sc., F.I.C.

CLARK has described^{1,2} the use of 4-methyl-1:2-dimercaptobenzene (dithiol) for the detection of tin; a reddish precipitate is produced on addition of this reagent to an acidified solution of a tin salt. In the course of some toxicological work I obtained with hydrogen sulphide a reddish-black precipitate which, when tested for tin by dissolving it in conc. hydrochloric acid and treating the solution in the manner described by Clark, yielded a greenish precipitate. Clark, in his paper, describes yellow, black and orange precipitates which are given by some of the more common metals with this reagent, but he does not mention a green precipitate. Further investigation showed this greenish precipitate to be due to the presence of molybdenum.

In view of the possibility that the formation of this greenish precipitate might be specific for molybdenum, salts of all the more common metals were tested with dithiol and, with the exception of tungsten, which produced a light greenish-blue precipitate, no other metal was found to yield a green precipitate.

ACTION OF AMMONIA ON THE METAL DITHIOL COMPLEX.—In the search for another metal that would produce a green dithiol complex, the procedure suggested by Clark, *viz.* the addition of thioglycollic acid and the dithiol reagent to a solution of the metal salt in dilute hydrochloric acid, was extended, and in each instance the effect of the addition of an excess of strong ammonia to the dithiol complex was examined. This additional procedure produced interesting results, in that it was found that all precipitates dissolved readily in an excess of ammonia, producing

nearly colourless solutions, with the exception of molybdenum, which produced a brilliant blue solution. This last reaction therefore gives us a specific test for molybdenum.

ACTION OF THIOLYCOLLIC ACID.—Another new colour reaction was also observed on addition of thioglycollic acid. Molybdenum ions, if present in the solution, produce a brilliant yellow colour; tungsten, on the other hand, yields no colour. Although this reaction with thioglycollic acid is not specific for molybdenum ions, it serves as a useful confirmatory test.

These two new colour reactions afford a rapid method for the detection of small quantities of either molybdenum or tungsten.

The tests give best results when the quantity of molybdenum or tungsten present in the 10 ml. of solution taken for examination is between 1.0 and 0.02 mg.

PROCEDURE.—Take 10 ml. of the solution to be examined and add sodium hydroxide solution or hydrochloric acid until the mixture is neutral to litmus, add 10 drops of conc. hydrochloric acid followed by 3 drops of thioglycollic acid, and observe the colour produced.

| Molybdenum | Tungsten |
|---------------|------------|
| Bright yellow | Colourless |

Add to this solution 1 ml. of the dimercaptobenzene reagent, prepared according to the direction of Clark (*loc. cit.*), boil for 3 or 4 minutes, and observe the colour of the precipitate.

| Molybdenum | Tungsten |
|------------------------|---------------------------------|
| Dark green precipitate | Light greenish-blue precipitate |

Cool the mixture and add an excess of 0.88 ammonia.

| Molybdenum | Tungsten |
|-------------|------------|
| Bright blue | Colourless |

By this test, 0.02 mg. of molybdenum in 10 ml. of solution may be detected and a similar quantity of tungsten will give a distinct greenish-blue dithiol complex.

When a suspension of the molybdenum complex in dilute hydrochloric acid is shaken with a mixture of equal parts of amyl alcohol and ethyl ether the complex dissolves in the ethereal mixture, with the production of an olive-green layer. The tungsten complex behaves similarly, yielding a blue layer.

The presence of traces of iron will interfere in the final stage of the test for molybdenum by producing on the addition of ammonia a violet colour with the thioglycollic acid present. Iron, if present, should therefore be removed by pouring the solution to be tested into an excess of sodium hydroxide solution, filtering off the precipitated ferric hydroxide, and testing the filtrate as previously described.

DETECTION OF TUNGSTEN IN PRESENCE OF MOLYBDENUM.—Tungsten may be detected satisfactorily, provided that the molybdenum is first removed by precipitation with hydrogen sulphide. The following experiment indicates the procedure that has been found quite satisfactory:—To 20 ml. of a solution containing 10 mg. of molybdenum and 0.1 mg. of tungsten, and made just alkaline to litmus by addition of sodium hydroxide solution, were added 2 g. of tartaric acid and 0.2 ml. of conc. sulphuric acid. The mixture was warmed to

60° C. and hydrogen sulphide gas was passed through it. The reddish-black precipitate was filtered off, the clear filtrate was evaporated to dryness, and the residue was gently ignited to destroy the tartaric acid. The residue was dissolved in a few drops of 20 per cent. sodium hydroxide solution, and the solution was diluted and examined by the method previously described. A distinct greenish-blue dithiol precipitate was obtained which dissolved in ammonia, giving a colourless solution, thus indicating the presence of tungsten and the absence of molybdenum.

It was found during the experiments on the separation of molybdenum and tungsten by this method that the presence of considerable quantities of tartaric acid or tartrates inhibited the dithiol reaction for tungsten; hence the necessity for destroying the tartaric acid before applying the test.

The method of storing the 4-methyl-1:2-dimercaptobenzene (dithiol) reagent suggested by Clark, *i.e.* in an atmosphere of hydrogen gas, has been found to be very satisfactory, and by this means the reagent may be kept for as long as 3 or 4 months without deterioration.

I wish to thank Dr. Bernard Dyer and Mr. George Taylor for their interest in this work.

REFERENCES

1. R. E. D. Clark, *ANALYST*, 1936, **61**, 242.
2. ——— *id.*, 1937, **62**, 661.

DR. BERNARD DYER'S LABORATORY

17, GREAT TOWER STREET, LONDON, E.C.3

November 4th, 1939

Bleach Ointments

BY D. D. MOIR, M.Sc., F.I.C.

IN view of the importance of bleaching powder preparations in the first-aid treatment of mustard gas casualties, the following notes and observations on the strength and keeping properties of bleach ointment may be of interest.

According to A.R.P. Handbook No. 2, 3rd edition ("*First Aid and Nursing for Gas Casualties*"), bleach ointment consists of equal parts of bleaching powder and white petroleum jelly. Handbook No. 3, 1st edition ("*Medical Treatment of Gas Casualties*"), specifies "supertropical" bleach. For either preparation it is reasonable to expect that the available chlorine in the bleaching powder should not fall below 30 per cent. (the B.P. limit); it is usually higher in powders stored under good conditions. Thus, bleach ointment, when freshly prepared, should contain about 15 per cent. of available chlorine. No specification is given for the white petroleum jelly in the Handbooks, although it is understood that there is an official Specification available to manufacturing chemists, but not generally published. The nature of the petroleum jelly used is, I believe, of the greatest importance. A yellow petroleum should not be used, since there may be interaction between the bleaching powder and the jelly, which can become very vigorous and has been known to cause a fire in large bulk. Even with white petroleum jelly this possibility is not entirely excluded.

The fact that bleach ointment is liable to deteriorate has been known for some time. Chambers and Savage,¹ who prepared bleach ointment according to the earlier A.R.P. formula of two-thirds bleaching powder and one-third petroleum jelly, observed a loss in available chlorine of about 50 per cent. in 15 months. Of this loss, a small proportion was found as combined chlorine in the petroleum base, but by far the greater part had been converted into inorganic chloride.

DETERMINATION OF AVAILABLE CHLORINE.—After several preliminary trials the following method of determining available chlorine was found to be most satisfactory and was used throughout these experiments:—About 1 g. of ointment is weighed directly into a weighed stoppered flask of about 200 ml. capacity. Twenty ml. of carbon tetrachloride are added, and the flask is rotated gently, with slight warming if necessary, until the whole of the paraffin is dissolved; 100 ml. of water are added and the flask is shaken vigorously. About 2 g. of solid potassium iodide are added, followed by 5 ml. of 33 per cent. v/v acetic acid. After a further shaking, the liberated iodine is titrated with *N*/10 sodium thio-sulphate solution. Vigorous shaking is sometimes necessary towards the end of the titration to decompose small particles of bleaching powder and to remove the iodine from the tetrachloride. Carbon tetrachloride was chosen because it is a good solvent for the petroleum jelly and is inert towards bleaching powder. The unsuitability of petroleum spirit was emphasised by Chambers and Savage, who showed that it was not without action on bleaching powder. I have not found any definite indication of loss of chlorine from the use of petroleum spirit, but the use of carbon tetrachloride avoids this possibility.

The petroleum base is determined by extraction with petroleum spirit in a Soxhlet thimble lined with filter paper; thus determined it would include any small quantity of chlorine which has entered into combination with the jelly.

I have examined about 40 bleach ointments, mainly taken from pharmacists in the ordinary course of Food and Drugs sampling, and two ointments have been prepared in the laboratory and assayed for available chlorine from time to time. The results are shown in Tables I to III.

Deficiency in available chlorine in a bleach ointment may be due to one or more of several causes. The wrong proportion of bleaching powder may have been used, or the bleaching powder may have been deficient in strength. Deterioration may have occurred either through interaction between the bleaching powder and unsaturated compounds in the petroleum base or by loss in strength of the bleaching powder itself. Any combination of these causes is possible, and it is not easy to distinguish between them.

ANALYSES OF BLEACH OINTMENTS.—The results of analysis can conveniently be classified under the following headings:

1. Ointments correctly prepared and of approximately correct strength (Table I).
2. Ointments deficient in available chlorine (Table II) owing to insufficient bleaching powder (Group 1), or to deterioration before or after preparation (Group 2), or to a combination of these causes (Group 3).

TABLE I

Available chlorine

| Sample No. | On receipt Per Cent. | After time stated in brackets Per Cent. |
|------------|-------------------------|---|
| B 103 | 14.7 | 14.4 (8 weeks) |
| | | 14.2 (17 ") |
| B 104 | 15.3 | 14.8 (8 ") |
| | | 14.8 (17 ") |
| B 107 | 12.9 | 11.6 (8 ") |
| | | 10.3 (17 ") |
| B 110 | 16.4 | 13.1 (17 ") |
| B 113 | 14.0 | 13.3 (17 ") |
| B 120 | 13.9 | 11.7 (16 ") |
| C 319 | 14.9 | 14.6 (16 ") |
| C 321 | 13.7 | 11.6 (16 ") |
| C 322 | 15.3 | 13.5 (16 ") |
| D 373 | 13.3 | 13.0 (15 ") |
| C 355 | 14.2 | 11.5 (13 ") |
| C 356 | 18.9 | 16.3 (13 ") |
| B 127 | 13.6 | 11.6 (12 ") |
| B 129 | 15.6 | 14.9 (12 ") |
| D 432 | 14.9 | 14.4 (12 ") |
| D 433 | 15.5 | 13.5 (12 ") |
| B 131 | 14.7 | 14.2 (12 ") |
| E 17 | 15.2 | 13.5 (11 ") |
| E 19 | 14.3 | 13.3 (11 ") |
| R 101 | 13.8 | 13.2 (9 ") |
| C.P. 24 | 13.7 | 13.4 (9 ") |

TABLE II

| | | Available chlorine | | Available chlorine expressed as percent- age of the bleaching powder present on receipt |
|------------|-------|----------------------------|--|---|
| Sample No. | | On receipt Per Cent. | After time stated in brackets Per Cent. | |
| Group 1 | B 109 | 62.3 | 10.8 | 8.7 (17 weeks) |
| | B 115 | 89.5 | 3.7 | 1.8 (17 ") |
| | B 121 | 93.4 | 2.3 | 1.3 (16 ") |
| Group 2 | B 114 | 52.1 | 11.5 | 7.5 (17 ") |
| | C 320 | 52.5 | 7.4 | 6.9 (16 ") |
| | C 353 | 52.5 | 7.7 | 6.3 (13 ") |
| | B 132 | 49.4 | 3.9 | 3.9 (12 ") |
| | B 133 | 45.6 | 8.3 | 7.7 (12 ") |
| | E 18 | 52.3 | 8.9 | 7.8 (11 ") |
| | R 95 | 50.5 | 10.6 | 9.3 (9 ") |
| | R 98 | 52.6 | 9.6 | 7.7 (9 ") |
| | R 100 | 48.8 | 8.1 | 4.9 (9 ") |
| | B 108 | 56.8 | 10.3 | 7.0 (17 ") |
| Group 3 | B 111 | 66.0 | 1.6 | 1.0 (17 ") |
| | B 112 | 67.9 | 8.2 | 5.7 (17 ") |
| | C 318 | 67.8 | 8.2 | 6.1 (16 ") |
| | D 372 | 57.0 | 7.7 | 6.2 (15 ") |
| | D 374 | 70.2 | 8.2 | 7.3 (15 ") |
| | C 354 | 56.9 | 11.0 | 10.3 (13 ") |

Of a total of 40 ointments examined, 21 were passed as satisfactory (Table I). The average available chlorine-content was 14·8 per cent. After periods of about 3 months, or more, the available chlorine had not in any instance fallen below 10·3 per cent. and averaged 13·3 per cent.

Samples Nos. B 104 and C 321 were from different vendors, but had been prepared by the same manufacturer; similarly with D 432 and E 17 and with C 319, B 129 and C.P. 24. It would seem, therefore, that manufacturers are able to prepare consistently bleach ointment of good strength and excellent keeping properties.

In Table II, B 115 and B 121 are informal and formal samples respectively from the same vendor. They were both prepared immediately before sale with a high-grade bleaching powder. The reduction in available chlorine on keeping is considerable, as is to be expected with such a low proportion of bleaching powder relative to the petroleum jelly.

The serious deficiencies in available chlorine of the samples listed in Table II, Group 2, which had the correct proportions of bleaching powder and petroleum jelly, can only be readily explained by assuming a long period of storage or the use of bleaching powder deficient in available chlorine. Several of the ointments were known to have been of recent preparation. The use of an unsuitable white petroleum jelly of high initial reactivity is not entirely ruled out, and is beyond the control of the vendor, who is unable to obtain the official Specification.

Sample No. R 100 gave on extraction with petroleum spirit a sticky residue instead of a dry powder. It was found to contain a considerable proportion of moisture, and it is probable that the bleach had been made into a paste before incorporation with the jelly.

TABLE III

LOSS OF AVAILABLE CHLORINE IN BLEACH OINTMENTS PREPARED IN THE LABORATORY.—No. 1.—Prepared with equal parts by weight of bleaching powder and white soft paraffin, both of which were taken from ordinary laboratory stock. The bleaching powder contained 30·0 per cent. of available chlorine, and the ointment should therefore have contained 15·0 per cent. exactly.

No. 2.—Prepared with equal parts by weight of bleaching powder and white petroleum jelly. The bleaching powder was taken from a freshly-opened tin direct from the suppliers, and the white petroleum jelly was also freshly purchased, but without any indication that it was suitable for use in this preparation. The bleaching powder contained 34·1 per cent. of available chlorine, and the ointment should therefore have contained 17·0 per cent.

| No. 1 | | | No. 2 | | |
|---------------------|----|------------------------------|----------------------|----|------------------------------|
| Time | | Available chlorine Per Cent. | Time | | Available chlorine Per Cent. |
| Immediately .. | .. | 14·2 | Immediately .. | .. | 16·5 |
| After one week .. | .. | 13·6 | After three weeks .. | .. | 13·3 |
| „ two weeks .. | .. | 13·2 | „ six weeks .. | .. | 11·9 |
| „ three weeks .. | .. | 12·7 | „ twelve weeks .. | .. | 11·6 |
| „ fifteen weeks .. | .. | 10·6 | „ fourteen weeks .. | .. | 11·5 |
| „ seventeen weeks.. | .. | 10·5 | | | |

At the end of these periods the bleaching powders with which the ointments were prepared contained 29·6 per cent. and 33·7 per cent., respectively, of available chlorine.

In both instances there was an immediate initial loss during preparation followed by a fairly rapid decrease in strength for the first few weeks. After about six weeks this loss became every slow.

Ointment No. 2 developed a yellow colour on keeping, although it was in the dark. As measured in the Lovibond Tintometer, the colour developed was equivalent to 1·6 yellow units in a period of two months. The figure was not increased after a further six weeks' storage. This phenomenon was unexpected in presence of available chlorine, but it was also observed in several of the ointments received under the Food and Drugs Act. Nos. B 108, B 115, B 121, B 127, B 132 and R 98, after storing, all had a slight yellow colour similar to that obtained with laboratory preparation No. 2. With one or two ointments which had been stored in cardboard boxes the yellow colour appeared to be more intense on the surface than underneath. This was particularly noticeable in B 126—an informal sample not listed above. The yellow colour developing on the surface of the ointment exposed to the air measured 8·0 yellow units after 3 months, whereas the formal sample, B 132, which was stored in an amber-glass jar with an air-tight screw cap did not exhibit this surface intensification.

These observations constitute a warning against condemning an ointment as having been prepared with yellow petroleum jelly instead of white, as judged by the colour alone. Nos. B 114, C 318 and E 18 were all strongly yellow in colour. No. E 18 was a sample taken from a large batch which had been supplied to a local authority over a year previously. It was believed to have been white when received.

Legal proceedings were taken against the vendors of samples Nos. B 111 (Table II, Group 3) and B 121 (Table II, Group 1), and reports appeared in *THE ANALYST*, 1940, p. 30. Both were extreme cases in which the available chlorine had reached such a low figure that the article was considered almost useless for its purpose. The cases were dismissed, but it is significant that the Chairman of the Bench at Sutton Police Court said "that a very useful purpose had been served by the case, for now the general public and druggists would know that something was advised in the air-raid precautions handbook which might be regarded as a standard and which it would be wise to keep to, at least until something more authoritative was found."

It is considered that a purchaser of bleach ointment for A.R.P. purposes is entitled to receive an article containing approximately 15 per cent. of available chlorine. It has been shown that in every instance when such an article has been sold the ointment has retained a high proportion of the active ingredient over a period of months and that subsequent deterioration is likely to be very slow.

REFERENCE

1. W. P. Chambers and F. M. Savage, *Pharm. J.*, 1938, **87**, 113.

ANALYTICAL LABORATORY
16, SOUTHWARK STREET
LONDON, S.E.1

January 24th, 1940

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

A SIMPLE TEST FOR ARSENIC IN LEAD ALLOYS

THE following limit test for arsenic in lead alloys, mainly those containing between 5 and 20 per cent. of antimony, has been devised in this laboratory. For this purpose the test has proved very useful, and, since it is simple and rapid, it may be of service in other connections.

Two g. of the alloy are heated in a 250-ml. conical flask with about 3 g. of anhydrous sodium sulphate and 15 ml. of conc. sulphuric acid, the contents of the flask being kept in motion to avoid local over-heating. The heating is best carried out over a large Bunsen flame by the use of flask tongs, and is continued until the lead salts appear almost white. After cooling, 75 ml. of water are added, sodium salts are dissolved by gently warming to the boiling-point, and the solution is treated with 75 ml. of strong hydrochloric acid, followed by 2 drops of saturated mercuric chloride solution and 3 ml. of hypophosphorous acid (sp.gr. 1.135). On heating the mixture to the boiling-point with frequent agitation, all lead salts dissolve, and, if arsenic is present in sufficient amount, a brown colloidal precipitate is produced.

As little as 0.005 per cent. of arsenic in the sample (0.1 mg.) gives a colour that is readily perceptible, and no interference is caused by 20 per cent. of antimony, or by any amounts of copper, silver, bismuth or tin that I have met with in antimonial lead. In absence of iron, which is seldom present, an amount of copper of the order of 0.03 per cent. or more is shown by the yellow colour of the warm hydrochloric acid solution, but the colour is discharged when hypophosphorous acid is added. The behaviour of selenium and tellurium is under investigation.

The precipitation of metallic arsenic by hypophosphorous acid in hydrochloric acid solution is well known,¹ but in the present test the sensitivity appears to be considerably enhanced by the small addition of mercuric chloride. Not more than 2 drops of the saturated solution should be used, as there is then likely to be some confusion with the grey colour of precipitated mercury.

Evans² used mercuric chloride to catalyse the reduction of tin by hypophosphorous acid, and it is from his paper that the suggestion of the present method arose. There is no doubt that mercuric chloride also considerably accelerates the reduction to metallic arsenic, the effect being more significant when the concentration of the latter element is small.

R. G. ROBINSON

REFERENCES

1. B. S. Evans, *ANALYST*, 1931, **56**, 523.
2. ——— *id.*, 1931, **56**, 170.

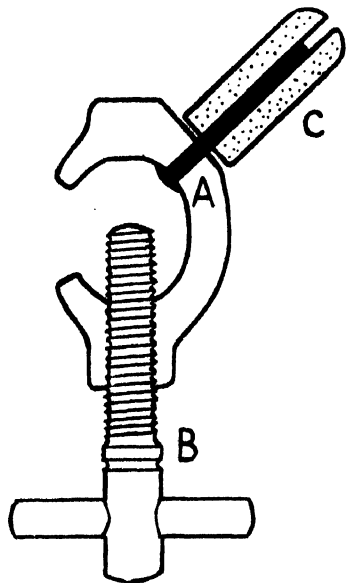
BRITANNIA LEAD CO.
NORTHFLEET

December 7th, 1939

AN ADJUSTABLE SUPPORT FOR CRUCIBLES AND DISHES

It often happens that platinum dishes and crucibles are heated on pipeclay or silica triangles even in laboratories where thermostatically controlled muffles are available, the muffles being reserved for certain types of work. The following support has been devised and possesses several distinct advantages.

The support is made from small retort bosses which are cut in half, three halves being required for each set. The halved retort boss is drilled with a $5/64$ inch hole at A at approximately 45° with the screw B. The boss is filed flat at A at the same angle. A $5/64$ inch wire or nail is thickened at one end and inserted from the inside where it is riveted in position. The wire is allowed to project $\frac{3}{4}$ inch and is fitted with a piece of pipeclay from a triangle or with silica tubing of similar size.



Three of these prepared attachments are fastened on the three sides of a triangular tripod with the screw B on the under side or pointing outwards, and with C pointing towards the centre and upwards. By fastening in the centre of the sides, the position is given for holding the smallest crucibles; when each is moved in the same direction along the side, *e.g.* to the left, the distance between the points can be increased as desired to take large dishes. The points should be arranged as far apart as practicable, so that the dish is held by its sides.

The device has the following advantages:—

(1) The crucible or dish is supported on three points and the increase in temperature is very appreciable. (2) The points can be readily removed for cleaning or renewal. (3) The device is easily adjustable and takes any size. (4) The crucible or dish is held rigidly and the stand can therefore be moved or rotated safely if desired. (5) The increase in efficiency means a saving in gas. (6) The support is cheap to make (about 1s. 6d.). (7) Where expense is of secondary importance platinum wire can replace the iron wire and can be used direct for supporting the crucible. The ends of the wire should then be bent to prevent denting the crucible.

I wish to thank Messrs. Cadbury Bros., Ltd., for permission to publish this note.

H. C. LOCKWOOD

CHEMISTS' DEPARTMENT
BOURNVILLE

September 25th, 1939

Official Appointments

THE following Notification of Amendment (dated December 16th) of the List of Public Analysts appointed by Local Authorities with the approval of the Minister of Health has been received from the Ministry.

| Authority | Name of Public Analyst |
|----------------------------------|-----------------------------------|
| NUNEATON BOROUGH | F. G. D. CHALMERS |
| HARROGATE BOROUGH | F. W. M. JAFFÉ |
| COULSDON AND PURLEY U.D. .. | E. HINKS |
| " " | D. D. MOIR (Deputy) |
| WIMBLEDON BOROUGH | E. HINKS |
| " " | D. D. MOIR (Deputy) |
| WINDSOR (NEW) BOROUGH | J. H. WEBER (Deputy) |
| DARTFORD BOROUGH | F. W. F. ARNAUD |
| CREWE BOROUGH | T. R. HODGSON |
| STRETTFORD BOROUGH | G. H. WALKER |
| BECKENHAM BOROUGH | J. W. FLINT (Deputy) |
| SWINDON BOROUGH | R. H. ELLIS |
| TORQUAY BOROUGH | THOMAS TICKLE |
| " " | C. V. REYNOLDS (Deputy) |
| BATTERSEA MET. BOROUGH .. | A. H. M. MUTER (Deputy) |
| MERTON AND MORDEN U.D. .. | E. HINKS |
| " " | D. D. MOIR (Deputy) |
| BROMLEY BOROUGH | J. W. FLINT (Deputy) |
| SCUNTHORPE BOROUGH | JOHN EVANS |
| WANSTEAD AND WOODFORD BOROUGH | { B. DYER } Joint Public Analysts |
| COLNE BOROUGH | { G. TAYLOR } |
| WIDNES BOROUGH | F. MAUDSLEY |
| | J. R. STUBBS |

ERRATUM.—In the list of Official Appointments on p. 27 of this volume, the last line, *viz.* Lincs. County, should be deleted.

Sugar for Chemical Purposes

PERMITS to purchase from a retailer sugar for chemical purposes may be obtained by application to the local Food Control Office. The appropriate form to be completed is numbered P.S.M.1.

Department of Scientific and Industrial Research

METHODS FOR THE DETECTION OF TOXIC GASES IN INDUSTRY*

ANILINE VAPOUR

OCCURRENCE.—Aniline exists in small quantities in coal tar, but is produced industrially almost exclusively by the reduction of nitrobenzene. It is used chiefly as a starting point in the manufacture of dyestuffs and to a less extent in other chemical industries. Its pleasant smell in no way suggests the dangers resulting from its presence in the atmosphere.

POISONOUS EFFECTS.—It has been shown that cases of acute aniline poisoning arise by absorption through the skin by splashes either directly on the skin or indirectly through the clothing. Its immediate toxic effect is on the blood, and the symptoms that characterise it are a blue-grey discoloration of the lips, ears and cheeks. Depending on the intensity of the concentration, shortness of breath, rapid feeble pulse, and nervous excitement, not unlike drunkenness, may occur later. With a concentration of 1 part in 140,000 of air, slight symptoms may occur after several hours' exposure, while with concentrations of 1 part in 10,000 to 1 part in 6000 serious disturbance occurs if the atmosphere is inhaled for more than an hour.

METHODS OF DETECTION.—In order to test for the presence of aniline vapour in the atmosphere, the aniline must first be brought into solution by drawing a sample of the air under examination by means of a hand-pump through a small bubbler containing dilute hydrochloric acid. Any aniline vapour present is thereby converted to the hydrochloride, to which any of the ordinary tests for aniline may then be applied.

Normally, however, the concentration of the vapour present in the atmosphere is so small that only very little hydrochloride is obtained without taking an inconveniently large sample of air; several of the ordinary tests are therefore insufficiently sensitive. For example, the best known test for aniline is by adding a few drops of a dilute solution of bleaching powder, which produces a purple colour, rapidly changing to a dirty red; over the range of concentrations of aniline vapour likely to be encountered in the atmosphere, however, only the slightest trace of colour is obtainable.

THE BLEACHING POWDER TEST.—A more sensitive test has therefore been developed from the bleaching powder test. If, after the addition of the bleaching powder, the solution is made alkaline with ammonia and a dilute aqueous solution of phenol is added, a permanent deep blue colour is produced, even with minute quantities of aniline. This test has been adopted as the standard test for aniline vapour in industry. It has been made quantitative by comparing the colours obtained at known concentrations with a series of standard colours prepared from a dye. In this way a table has been drawn up showing the depth of colour obtained with up to 10 strokes of the standard hand-pump over a range of concentrations from 1 part in 5000 to 1 part in 100,000. The table is included in the Leaflet with full instructions for making the tests and for the preparation of the standard colours.

* Leaflet No. 11. H.M. Stationery Office, York House, Kingsway, London, W.C.2. December, 1939. Price 3d. net.

FOREST PRODUCTS RESEARCH BOARD

REPORT FOR THE YEAR 1938*

AN outline is given of the activities of the Laboratory in investigational, educational and advisory work.

IDENTIFICATION OF TIMBER.—About half of the numerous enquiries were concerned with the identification of timber. A representative collection of specimens of softwoods has been made, and from the examination of all characteristics of diagnostic value a multiple-entry card-key (illustrated in the Report) has been devised.

EFFECT OF CHANGES OF MOISTURE-CONTENT OF WOOD.—Investigations of the methods of treating timber to prevent swelling or contraction through changes in its moisture-content have been made. So far, the best results have been obtained by soaking the wood in a solution of sorbitol. In experiments with green beech boards ($\frac{1}{2}$ in. thick) it was found that movement was appreciably reduced when a fairly high concentration (15 to 20 per cent. by weight) of sorbitol was present. The results obtained with oak boards (1 in. thick) were still better. The presence of about 40 to 50 per cent. (by weight) of sorbitol reduces the movement by 40 to 50 per cent.

ABSORPTION OF MOISTURE.—The rates of absorption of moisture by oak, mahogany, teak, red deal, white deal and British Columbian pine, when stored in a damp situation (25° C., 90 per cent. humidity), are given in the form of a graph.

When dry timber is stored for long periods, or alternatively when air-dried timber is to be further seasoned to make it suitable for indoor use, it is very desirable that the storage conditions should correspond, as far as possible, to a moisture-content of about 12 per cent. In a climate with high humidity, as in this country, the required conditions can generally be attained by slight heating. A simple apparatus has been devised for automatically controlling the air conditions in a store. A block of wood is first conditioned to the desired moisture-content and adjusted so that its expansion, as a result of increase of the moisture in the block, closes a switch which brings the heating apparatus into play. The rise in air temperature lowers the humidity, thus causing the wood block to dry and shrink and so break the electric contact at the switch.

FIRE RESISTANCE OF TIMBER.—More than 70 different species have been tested by the laboratory standard methods of determining inflammability, resistance to flame penetration and rate of burning. Those showing outstanding resistance to fire were: Greenheart (*Ocotea rodioei*), gurjun (*Dipterocarpus* spp.), jarrah (*Eucalyptus marginata*), laurel (*Terminalia tomentosa*), padauk (*Pterocarpus macrocarpus*), pyinkado (*Xylia dolabriformis*), teak (*Tectonea grandis*), and white olivier (*Terminalia obovata*).

ACTION OF FIRE-PROOFING COMPOUNDS.—Previous work has shown that the main action of a fire-retarding chemical is its influence on the heated wood, which results in the formation of charcoal at the expense of the production of inflammable gaseous compounds. Further work has confirmed the conclusion that the most effective chemicals are those dissociated by heat, with liberation of an acid radical. Thus, the products from the heating of wood at 450° C. contained 26 per cent. of charcoal, 33 per cent. of condensable organic vapours and 19 per cent. of water; those from the wood after treatment with a 20 per cent. solution of ammonium chloride contained: charcoal, 48; condensable organic vapours, 9; water, 35 per cent. The increase in water vapour also has an effect.

A NEW PRESERVATIVE MIXTURE.—Laboratory tests have indicated that the presence of potassium or sodium dichromate often enhances the leaching resistance of water-borne toxic preservatives. Arising from these tests a new wood preservative mixture has been devised. The addition of potassium dichromate to

* H.M. Stationery Office, York House, Kingsway, London, W.C.2. 1940. Price 1s. 6d. net.

mercuric chloride solution resulted in the fixation of the mercury in the wood; under severe test conditions less than 5 per cent. of the mercury was leached out of the treated wood.

CONTROL OF *LYCTUS*.—The principal work on the control of *Lyctus* (powder-post beetles) has been concerned with the investigation of methods of reducing the starch-content of timber.

Estimation of Starch in Timber.—The standard method adopted consisted in treating a clean-cut radial longitudinal surface with a 0.5 per cent. solution of iodine in potassium iodide and estimating visually the amount of starch in each 1/10 inch of wood from the outside of the tree inwards as falling into one of an arbitrary series of grades.

DETECTION OF WOOD-BORING INSECTS BY MEANS OF X-RAYS.—It has been found that, although the presence of insect tunnels can be discovered by X-ray examination, the method is likely to be restricted to timber of small dimensions, such as plywood. It has not proved successful for detecting the death-watch beetle and its larvae in structural timbers.

DESTRUCTION OF INSECTS IN WOOD BY FUMIGATION.—In test experiments exposure, in a commercial fumigation chamber, of the infested wood to the vapours of hydrogen cyanide for 4½ hours at 70° F. killed most, if not all, of the larvae.

PHYSIOLOGICAL STUDIES OF WOOD-DESTROYING FUNGI.—Those fungi that produce a brown rot (*i.e.* attack cellulose and carbohydrates only) cause the medium to become and remain very acid, whilst those that produce a white rot cause an initial development of acidity which disappears after some weeks. The reason for this difference is under investigation.

OTHER INVESTIGATIONS.—Other work described in detail in the Report includes the successful manufacture of charcoal in portable kilns, the effect of repeated applications of oil to wood flooring, the drying of timber by high frequency electric fields such as those produced by radio transmitters, box-testing, the application of statistical analysis to wood bending, and vibration methods for measuring the elastic constants of wood.

Queensland

ANNUAL REPORT OF THE GOVERNMENT ANALYST TO JUNE 30TH, 1939

IN his Annual Report Mr. F. E. Connah, F.I.C., gives a summary of the work done for various departments. The 14,911 samples analysed included 7510 for the Health Department, 2616 for Customs, 1404 for the Postmaster, and 1081 for the Geological Survey. Of the samples for the Health Department, 3349 were taken by inspectors under the Health Act; 2841 of these were passed as satisfactory.

LEAD IN CRAYONS.—Of the 1018 samples examined, 145 were rejected. The amounts of lead in legal samples were: Green (23), 0.1 to 20.4 (aver. 4.0); yellow (19), 0.1 to 14.0 (aver. 2.6); orange (7), 0.2 to 13.0 (aver. 5.4); brown (4), 1.4 to 6.0 (aver. 2.6); red 0.2 to 2.0 (aver. 0.8) per cent. Some black crayons also contained lead (0.9 to 2 per cent.). Most of the lead was soluble in 0.25 per cent. hydrochloric acid.

PARAFFIN OIL ON CURRANTS AND RAISINS.—Seven samples of currants (average water-content 18 per cent.) yielded 0.04 to 0.2 per cent. of ethereal extract. Raisins (7 samples with average water-content 17 per cent.) yielded 0.06 to 0.4 per cent.; 21 samples of sultanas (average water-content 16 per cent.) yielded 0.08 to 0.6 per cent. Any excess of ethereal extract over 0.1 per cent. may be accepted as due to paraffin oil. It is suggested that 0.1 per cent. might be taken as a desirable limit.

COMPOSITION OF "SPECIAL" COFFEES.—Ten samples of "special" coffees sold by local coffee inns gave the following results:

| | Mocha | Kenya | Boengi | Java | Costa Rica | Santos | Mysore | Queensland | Columbia | Blue Mountain Jamaica |
|---|-----------|-----------|-----------|-----------|------------|-----------|-----------|------------|-----------|-----------------------|
| | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. |
| Moisture .. | 3.4 | 3.4 | 3.2 | 3.4 | 3.7 | 3.8 | 2.6 | 2.8 | 2.6 | 3.0 |
| Caffeine .. | 1.1 | 1.2 | 1.2 | 1.8 | 1.3 | 1.3 | 1.2 | 1.1 | 1.2 | 1.1 |
| Ethereal extract | 14.0 | 15.2 | 14.3 | 10.8 | 14.2 | 14.5 | 13.0 | 15.0 | 13.0 | 11.9 |
| Crude fibre .. | 13.2 | 12.4 | 12.0 | 12.3 | 12.4 | 12.7 | 13.2 | 13.6 | 12.7 | 13.4 |
| Total ash .. | 4.6 | 4.6 | 4.5 | 4.6 | 4.7 | 4.7 | 4.9 | 4.6 | 4.9 | 5.0 |
| Water-soluble ash .. | 3.6 | 3.6 | 3.3 | 3.5 | 3.7 | 3.4 | 3.6 | 3.5 | 3.7 | 3.7 |
| Alkalinity of water-soluble ash (as K_2CO_3) | 2.8 | 2.9 | 2.6 | 2.7 | 2.9 | 2.9 | 3.1 | 3.0 | 3.1 | 3.2 |
| Cold-water extract .. | 21.2 | 20.8 | 20.8 | 21.4 | 21.7 | 21.1 | 19.6 | 18.2 | 21.0 | 20.5 |

The sp.gr. of the cold-water extract at 15.5° C. ranged from 1.0093 to 1.0105, the average being 1.0097. Whilst these coffees were very similar to ordinary coffee in chemical characteristics, some of them showed pronounced differences in aroma and flavour.

AMMONIA IN TOILET PREPARATIONS.—Ten samples used in the non-electric method of hair-waving consisted of aqueous solutions of ammonia and sodium sulphides. The ammonia-content ranged from 3.9 to 9 per cent. and six samples contained more than 5 per cent., thus coming within the scope of the Poisons Regulations. The sulphide (as hydrogen sulphide) ranged from 0.45 to 2 per cent. A solution containing 9 per cent. of ammonia would reduce hair strength to a pronounced extent, but the effect of 5 per cent. solutions would probably be transitory. In considering the question of operatives using such solutions it was calculated that if an ounce of solution (sufficient to treat the hair of two persons) containing 9 per cent. of ammonia and 2 per cent. of hydrogen sulphide were completely evaporated in a room, 12 ft. square and 12 ft. high, the concentration of ammonia in the atmosphere would be 0.007 per cent. and that of hydrogen sulphide 0.001 per cent. Such concentrations, according to authority, are non-poisonous, though objectionable in odour. The process is noxious and, if not unhealthy, certainly vitiates the air.

PARAFFIN AS "MACASSAR" OIL.—There is no evidence available that genuine macassar oil has any beneficial effect on the hair or prevents baldness. The hair oil described as "Macassar" in Queensland was found to consist almost entirely of paraffin oil. To claim that this oil is a "scalp food" suggests colossal ignorance of human metabolic processes.

LEAD IN BONE.—Fifty-six specimens of human skull and rib-bone were examined for lead by a wet oxidation process followed by extraction and estimation with dithizone. The specimens included both normal and nephritic cases. In no instance was a quantity of less than 1 mg. of lead per 100 g. of material found. The skull contained about twice the proportion found in the corresponding rib.

TRANSPORT OF "SODA CRYSTALS."—Ordinary washing soda (37 per cent. of Na_2CO_3) begins to "weep" at 90° F. and at 95° F. liquefies and causes damage to adjacent goods. These temperatures are often reached in Queensland, and the fact that more serious damage is not often reported is probably due to the chilling effect as the water of crystallisation is thrown off. Neither soda ash nor "bath salts" ("crystal carbonate") exude water in this way; they might be used as substitutes for "soda crystals."

Palestine

REPORT OF THE GOVERNMENT ANALYST FOR THE YEAR 1938

THE chemical work supervised by the Government Analyst (Mr. G. W. Baker, F.I.C.) is summarised in a special section of the Annual Report of the Department of Health. The number of samples examined under the Food Ordinance Rules was 8738, and 454 of these were adulterated. The principal foods examined were raw milk 4945 (137 adulterated), butter 565 (78 adulterated), semni 1547 (39 adulterated), olive and other oils 698 (114 adulterated), coffee 178 (13 adulterated).

JAM.—The Ordinance defines jam as some kind of fruit or fruits with sugar. The kinds of fruit must be stated on the label, and the addition of pectin, fruit juice and glucose is permitted. The cheaper jams are little more than pectin jelly coloured and flavoured. Of the 68 samples examined, 27 were returned as below standard.

ARTIFICIAL HONEY.—Various kinds of artificial so-called honey were examined. Some were cane or glucose syrups, whilst others were imitations of genuine honey made from invert sugar to which a honey flavour had been added. The colour tests for artificially inverted sugar did not always give positive results with these products.

APPLICATION OF BEAM'S TEST FOR HASHISH.—In one case in which a "nargileh" (hubble-bubble pipe) had been submitted it was possible to get a good result by applying Beam's test for hashish to the petroleum spirit washings from the bowl and stem of the pipe.

EXAMINATION OF BURNT CURRENCY NOTES.—In a claim for payment in connection with the charred remains of alleged currency the inscriptions on many of the notes could be identified by completing the ashing process (*cf.* Mitchell, ANALYST, 1925, 50, 174), and after prolonged search 27 different numbers on notes were read and recorded.

ARSENICAL INSECTICIDES AND WINE.—It has been noted that the use of arsenical insecticides in the vineyards may result in contamination of the wine. In some instances such wines have been successfully treated with iron oxide.

NEUTRAL SALTS AND SOILS.—In a special investigation it was found that the observation that the pH of soil is lowered by the addition of a neutral salt is valid for arid soils as well as for humid. All the processes that render humid soils unsaturated take place also in arid climates, but to a less extent.

Experiments on the effect of saline water on the "loess" type soils of the Beersheba area showed that under laboratory conditions there was little accumulation of salt by evaporation, but base exchange produced alkalinity. The soil was highly calcareous and the water used was very saline, containing 300 parts of chlorine per 100,000.

New Zealand Pharmacy Act

DEFINITION OF A DRUG

THE New Zealand Pharmacy Act, 1939, came into force on January 1st. The Act, which amends the 1908 Act, gives power to the Board of the Pharmaceutical Society of New Zealand, to make rules *inter alia* to promote proper conduct among pharmacists, such as regulating, advertising and prohibiting the use of specified methods of selling drugs. Membership of the Society is compulsory for pharmacists.

In the Act "drug" means any drug (as described in any official pharmaceutical publication) used in the treatment, prevention, investigation or alleviation of any

disease, illness or injury affecting human beings. "Official pharmaceutical publication" means the latest edition for the time being of the British Pharmacopoeia or of the British Pharmaceutical Codex, and includes any supplement or addendum to either of the said publications.

The First Schedule of the Act gives a list of articles the sale of which is uncontrolled. These include acetylsalicylic acid tablets, camphorated oil, cascara, malt and oil, senna leaves, etc. Other B.P. and B.P.C. preparations may be sold only by pharmacists.

British Pharmacopoeia Commission

REPORT OF THE COMMITTEE ON GENERAL CHEMISTRY*

THIS report has been published in order to provide the opportunity for medical practitioners, pharmacists, analysts, manufacturers and others who may be interested, to criticise it and to suggest emendations before the preparation of the new Pharmacopoeia reaches its final stages.

SECTION I.

The Sub-Committee on Alkaloids and Alkaloidal Salts have reviewed the monographs submitted to them, in so far as the descriptions, characters and tests for identity and for purity are concerned. They recommend certain changes (mainly concerned with the melting-points) in nine monographs.

SECTION II.

The Sub-Committee on General Organic Chemicals recommend changes or additional requirements in 19 monographs.

ÆTHER.—The Sub-Committee recommend that the monograph on Anaesthetic Ether in the current Pharmacopoeia should be continued, and the quality now described as "Æther" should be named "Æther Solvens, Solvent Ether."

GLYCERINUM.—The refractive index should read "1.4696 to 1.4726" instead of "1.470 to 1.472." A new test for reducing substances is proposed.

UREA.—The synonym "Carbamide" should be inserted. The m.p. (130°–132°) should be given as a Test for Purity.

SECTION III.

The Sub-Committee on Inorganic Chemicals recommend changes or additional requirements in 42 monographs and in Appendix I.

APPENDIX VI.—For the *Quantitative Determination of Lead* the Sub-Committee recommend the adoption of the ether-extraction method, and the necessary additions to the table on pp. 553 to 558 of the B.P., 1932, are given. These include the following limits for lead in p.p.m. Acidum Mandelicum, 5; Calcii Mandelas, 5; Ferri Carbonas Saccharatus, 50; Ferri et Ammonii Citras, 50; Ferri et Quininae Citras, 50; Ferri Subchloridum Citratum, 75; Ferri Sulphas, 30; Ferri Sulphas Exsiccatus, 50; Liquor Ferri Perchloridi, 15; Magnesii Sulphas Exsiccatus, 10; Potassii Chloridum, 5; Sodii Metabisulphis, 20; Sodii Phosphas Exsiccatus, 10; Sodii Sulphas Exsiccatus, 10.

APPENDIX VII.—*Quantitative Test for Arsenic.*—Additions are required in relation to certain substances which are to be added to the Pharmacopoeia. Amounts to be taken for the test are prescribed, and the following limits (p.p.m.) for arsenic are recommended:—Acidum Mandelicum, 2; Bismuthi Subgallas, 2; Calcii Laevulas, 2; Calcii Mandelas, 2; Magnesii Sulphas Exsiccatus, 5; Potassii Chloridum, 1; Sodii Metabisulphis, 5; Sodii Phosphas Exsiccatus, 10; Sodii Sulphas Exsiccatus, 4.

APPENDIX VIII. C.—*Limit Test for Iron.*—A colorimetric method based on the use of ammonium thiocyanate is described. The comparison is made in Nessler cylinders.

APPENDIX V.—*Qualitative Reactions and Tests for Substances mentioned in the Pharmacopoeia Tests*, regarded as specific, are given for 22 metals or acid radicals. Eight additional Reagent Solutions are proposed.

* Report No. 14. Published by authority of the General Medical Council, 44, Hallam Street, London, W.1. December, 1939. Price 1s. 6d.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Determination of Sugars in Flours. W. Iwanowski and G. Grabouska. (*Ann. des Fermentations*, 1938, p. 432; *J. Pharm. Belg.*, 1940, **22**, 24.)—The action of diastase and micro-organisms during the determination of sugars in flours can be prevented by using an alkali solution of exactly controlled concentration. The authors recommend a mixture of 75 per cent. of 0.15 *N* sodium carbonate solution, 25 per cent. of 0.15 *N* sodium bicarbonate solution and a few drops of chloroform. With this solution no decomposition of the sugars, starch or proteins occurs.

E. M. P.

Determination of Formic Acid in Foodstuffs. J. Grossfeld and R. Payfer. (*Z. Unters. Lebensm.*, 1939, **78**, 1–30.)—It is known that formic acid is not completely volatile in steam, owing to the formation of a hydrate, and the present work is concerned with its volatility in the vapour of organic solvents that are insoluble in water. It was found that no decomposition of formic acid took place when it was boiled for several hours in toluene, benzene or petroleum spirit, but only by a prolonged distillation-period or a large excess of solvent could formic acid be completely distilled with benzene or light petroleum spirit. On the other hand, a yield of over 95 per cent. was obtained from a mixture of 6 ml. of a solution of the acid in water and sufficient toluene or benzene (b.p. 100° to 110° C.) to produce distillates of 100 or 200 ml., respectively. Similar results were obtained with acetic and propionic acids in toluene, but the yields of lactic and butyric acids were less, and those of benzoic and valeric acids were still poorer; the yield of nonylic acid was only about 2.4 per cent. Prolonged distillation with toluene or benzene in presence of phosphoric acid results in the caramelisation of sugars, and the consequent production of formic acid and allied substances. The determination of formic acid by means of bromoacetic acid is unsatisfactory when the solutions are very dilute, and errors may be caused by salicylic, malonic, oxalic, lactic or tannic acid. Caramelisation of sugar is minimised if only 60 ml. of toluene or 120 ml. of benzene are used, although the yield of formic acid is then only 75 per cent.; losses may occur if the distillation is carried out too rapidly. "Interrupted distillation," in which the distillation is carried out in a special apparatus in stages, with addition of water whenever caramelisation starts, is therefore adopted (see below) as a compromise between the above limitations. In this way yields exceeding 97 per cent. can be obtained, except when the quantity of formic acid present is large. Where the quantity of formic acid is very small in relationship to the sugars present, it is desirable first to separate the former before distillation by means of a "perforation" process in a special apparatus (see below), lime water being used to fix the free acid. This, in turn, necessitates a solution that is quite free from colloidal substances, and a modification of the Carrez method of clearing is described, which does not involve the risk of the formation of ammonium formate by the hydrolysis of hydrocyanic acid. Inversion of sucrose with heat produces small quantities of formic acid detectable by the method

described, most of the reducing substances formed being left in the residue after distillation; inversion at room-temperature produces less of the interfering substances. The method finally recommended is as follows:—A solution of the sample (*e.g.* 125 g. of honey), which should not contain more than 100 mg. of formic acid, is diluted to 150 ml. in a 250-ml. graduated flask and acidified with 20 ml. of sulphuric acid (1 + 3). It is then cleared by the addition of 5 ml. of a 15 per cent.* solution of potassium ferrocyanide, followed, after shaking, by 5 ml. of 30 per cent.* zinc sulphate solution; the mixture is then made up to 250 ml., shaken, and filtered on a dry folded paper on which has been sprinkled a little kieselguhr. Clearing has been satisfactory if on shaking equal volumes of the filtrate and ether in a test-tube, no emulsion forms at the interface. If an emulsion does form, 200 ml. of the filtrate are treated with 25 ml. of a solution containing 60 g. of sodium phosphate and 100 g. of sodium tungstate per litre, and the mixture is diluted to 250 ml. and filtered on the next day through a dry folded paper; in many cases this latter stage of clarification is unnecessary. The "perforation" process then follows, 200 ml. of the filtrate being placed in the apparatus, which consists of a narrow vertical container (capacity, 400 ml.) fitted with a reflux-condenser and a side-arm just above its mid-point. Immediately under the lower end of the condenser is a narrow vertical tube, which is placed centrally in the container; this widens out slightly at the top, and at the bottom, where it rests on the base of the container, it is bent upwards and terminates in a horizontal sintered glass plate (G.1). The side-arm delivers into a conical flask which contains 0.5 g. of calcium oxide and 50 ml. of ether, and sufficient ether is added to the container to form on top of the aqueous solution a layer which just overflows into the side-arm without allowing any of the water layer to do so; all joints are of ground glass. The container is heated so that the ethereal layer in it evaporates, condensate being returned by the reflux condenser to the central tube, down which it passes and leaves as a stream of fine droplets through the sintered plate. These droplets pass up through the water layer to supplement the ethereal layer above it, and the excess of ether overflows through the side-arm. This operation lasts 5 to 6 hours, after which the distillate is evaporated and the mixture of calcium salts of organic acids and calcium hydroxide which remains is extracted for 5 minutes with 15 ml. of water, to which is added a little decolorising charcoal. The mixture is then filtered and the filtrate and washings are evaporated in presence of a little pumice powder, 5 ml. of water and sufficient 25 per cent. phosphoric acid (sp.gr. 1.154) to liberate the acids (*e.g.* 1 ml.) being added to the residue. This mixture is then distilled with 150 ml. of benzene (b.p. 80° to 90° C.) in a Payfer apparatus, the receiver of which is a wide burette-tube arranged vertically with the tap at the lower end, and joined at the top to a wider tube which is fitted with a reflux condenser. At the point where the tube widens, a side-arm (in the top end of which is inserted the outlet from a small graduated tap-funnel) leaves at a downwards angle, and terminates in a conical flask fitted with a ground-glass joint, which contains the mixture to be distilled. In this way a measured quantity of water may be added through the tap-funnel during the distillation ("interrupted distillation," see above), and the volume of benzene distilled and the distillate may be

* g. per 100 ml.

measured and easily removed from the receiver. The same conical flask used as a receiver in the "perforation" process can conveniently serve subsequently as the distillation flask. Distillation is carried out at such a rate that the distillate just falls from the condenser in a continuous stream, and when 3.5 ml. of aqueous distillate have been collected a further 5 ml. of distilled water are added to the distillation flask from the tap-funnel. When this, too, has distilled over 5 ml. more of water are added, and so on, until an aqueous distillate of at least 19.5 ml. is obtained; this takes about 45 minutes. The aqueous portion of the distillate is drawn off about 10 minutes after the conclusion of the distillation and evaporated, together with the washings from the apparatus, in presence of 1 g. of calcium carbonate. The dry residue is extracted with water, the extract is filtered, and the residue is washed until the filtrate amounts to 50 ml. The filtrate is then heated for an hour in a boiling water-bath with 15 ml. of a solution prepared by heating 10 g. of mercuric chloride, 4 g. of sodium chloride and 10 g. of crystalline sodium acetate with 100 ml. of water; for 1 hour on the water-bath it is cooled and filtered (cf. C. Zäch, *Mitt. Lebensm. Unters.*, 1933, **24**, 35); 15 ml. of this solution will oxidise up to 127 mg. of formic acid. After the oxidation the solution is filtered on a weighed glass crucible (No. 10G.3), and the precipitate is dried and weighed; 1 g. of Hg_2Cl_2 weighed \equiv 97.2 mg. of formic acid. The benzine may be recovered for re-use by extracting it with a quarter of its volume of water, then distilling it with sodium hydroxide solution, and finally clearing it with kieselguhr and filtering. The "perforation" process is essential with honey if its sugar-content is more than 100 times its formic acid content. In presence of alcohol the sample should be shaken with calcium carbonate until neutral, and the alcohol then removed by evaporating the mixture on the water-bath to half its volume. The solution is then decolorised with activated carbon, filtered and distilled as described. Values found were as follows:—Natural blossom honeys (4 samples), 2.9 to 5.8; leaf honey, 20.9; artificial honey, 18.6; cherry juice (preserved with formic acid), 218.5 and 242.5; raspberry juice (preserved with formic acid), 293.9; currant juice, 6.6; blood-orange juice, 2.8; sweet-orange juice, 2.8; maple syrup, 30.4; "food syrup," 23.4 mg. of formic acid per 100 g. J. G.

The Bellier Test and the Blarez Test. Application to Olive and other Oils. R. Marcille. (*Ann. Chim. anal.*, 1939, **21**, 311–321.)—The technique of the Bellier test is described in detail. The following values for different oils were obtained:—arachis, 38–41; coconut, 10.5; cod-liver, 9.5; mastic tree oil, 16.75–17.75; linseed, 16.5–20; olive, 9.5–19; castor, 0.5; soya bean, 16–19; tea seed, 7–10. The following results were obtained with olive oils of different origin:—Morocco, 10.5; Tunis, 16; Tunis + 10 per cent. of arachis oil, 19; Sfax, 17; Sfax + 10 per cent. of arachis oil, 20.5. Very acid olive oils, oils from fermented olives and marc oils, even when refined, tend to give precipitates attributed to wax-like substances, and with such oils the tube should be left for some time at 22–25° C.

Bellier Values of Solid Fatty Acids.—The determination of the Bellier values of mixtures of solid fatty acids gives results more definite than are obtainable by titration or determination of the melting-point. Thus, mixtures of pure palmitic and stearic acids gave the following results:

| | | | | | | | | | |
|-------------------------|------|------|------|-------|------|------|------|------|------|
| Stearic acid, per cent. | 0 | 10 | 20 | 30 | 40 | 50 | 60 | 75 | 100 |
| Melting-point °C. .. | 62 | 60.1 | 57.5 | 55.1 | 56.3 | 56.6 | 60.3 | 64.1 | 69.2 |
| Bellier value .. | 19.5 | 22 | 23.5 | 24.75 | 26 | 27 | 28 | 29.5 | 33.5 |

Similarly with mixtures of palmitic and arachidic acids:

| | | | | | |
|---------------------------|------|------|----|----|------|
| Arachidic acid, per cent. | 0 | 10 | 20 | 50 | 100 |
| Melting-point, °C. .. | 61.5 | 56.5 | 55 | 61 | 73.5 |
| Bellier value .. | 19.5 | 36.5 | 44 | 53 | 60 |

Special applications of the test are discussed, such as its application to the detection of the adulteration of arachis oil with linseed oil.

Examination of the Oil from Canned Fish.—Sardine oils have a Bellier value ranging from 18 to 19.5 and tunny-fish oil 19.5 to 21. It is exceptional for olive oil in boxes of sardines to show a value higher than 15.75, whilst olive oil adulterated with 10 per cent. of arachis oil has not given a value higher than 19.5.

The Blarez Value.—The temperature of crystallisation of the alcoholic solution of the potassium soaps of the fatty acids obtained by saponification of 1.5 ml. of oil with 15 ml. of alcoholic potassium hydroxide (45 g. per litre) is called the Blarez value. Pure olive oils give figures between 10 and 16 with an average of 12, and the addition of 10 per cent. of arachis oil raises the figure to 16.5; of 20 per cent. to 18.75; of 30 to 21; of 50 to 25.5, whilst the value for pure arachis oil is 28. Olive oils from canned fish may show abnormal figures and the results should be controlled by determining the Bellier values. D. G. H.

Fatty Acids and Glycerides of the Oil from Sapota Seeds. N. L. Vidyarthi and V. Mallya. (*J Indian Chem. Soc.*, 1939, 16, 443–448).—The evergreen sapota trees (*Achras sapota*, N.O. *Sapotaceae*), grown in southern India, produce edible fruits containing 2 or 3 seeds which are wasted. The easily removable hard shells of the seeds enclose the kernels (50 per cent. of the seed), which contain about 20 per cent. of light-coloured oil extractable with carbon tetrachloride. A sample of the oil had the following characteristics: sp.gr. at 31° C., 0.8725; n_D^{31} , 1.463; saponification value, 205.4; iodine value (Wijs), 59.8; Reichert–Meissl value, 2.8; Hehner value, 92.6; acid value, 8.94 per cent. (as oleic acid); unsaponifiable matter, 1.8 per cent. The component acids of the oil were: lauric, 1.6; myristic, 6.2; palmitic, 12.6; stearic, 12.0; oleic, 66.2; linolic, 1.4 per cent. The glycerides were calculated from data obtained by oxidising the oil with potassium permanganate in acetone, and by systematic crystallisation from acetone, and were: oleo-palmitostearin, 5; dioleomyristin, 23; dioleopalmitin, 36; dioleostearin, 28; triolein, 5; with 3 per cent. of disaturated mono-olein, in which the saturated acids are probably lauric, myristic and a little palmitic. No fully saturated glycerides were present. D. G. H.

Fatty Acids and Glycerides of the Fat from the Seeds of *Garcinia indica* (Kokum butter). N. L. Vidyarthi and C. J. Dasa Rao. (*J. Indian Chem. Soc.*, 1939, 16, 437–442).—The seeds from *Garcinia indica* trees (N.O. *Guttiferae*), growing on the western coast of the Madras and Bombay Presidencies, contain 20 to 25 per cent. of kokum butter, used for edible and medicinal purposes. The fat extracted from the crushed seeds with carbon tetrachloride had the

following characteristics:—sp.gr. at 40° C., 0.899; n_D^{40} , 1.4571; m.p., 39.4° C.; saponification value, 189.2; iodine value (Wijs), 36.7; free fatty acids (as oleic), 7.8 per cent.; unsaponifiable matter, 1.2 per cent. By the lead-salt separation and ester fractionation methods the fatty acids were found to consist of: myristic, 1.2; palmitic, 5.3; stearic, 52.0; oleic, 41.5 per cent. The neutral fat was oxidised in acetone and also submitted to systematic crystallisation from acetone, whereby two fractions were obtained, which were hydrogenated. The component glycerides were then calculated as: tristearin, 2.0; oleodistearin, 59; dioleostearin, 21; oleopalmitostearin, 14; oleodipalmitin, 2; palmitodiolein, 2 per cent. Compared with cocoa butter and Borneo tallow the fat contains a very small proportion of palmitic acid, but the high proportion of oleo-distearin tends to make the fat harder than those.

D. G. H.

New Zealand Fish Oils. III. Composition of the Depôt Fats of the Ling (*Genypterus blacodes*). F. B. Shorland. (*Biochem. J.*, 1939, 33, 1935–1941.)—Analyses of 7 samples of ling-liver oil taken at different periods showed no significant variation in fatty acid composition, which was in general similar to the "average" marine type with increased proportions of C_{18} unsaturated acids. The accuracy of the ester fractionation procedure was tested by various methods, all of which gave similar values for the proportion of component fatty acids. The following are typical results for the composition of the fatty acids (weight per cent.) of oils from various organs, the figures in brackets representing the degree of unsaturation:

| | Saturated | | | Unsaturated | | | |
|-------------------|-----------|----------|----------|--------------|---------------|---------------|----------------|
| | C_{14} | C_{16} | C_{18} | C_{16} | C_{18} | C_{20} | C_{22} |
| Liver | 1.9 | 16.9 | 2.6 | 6.5 (2.0) | 34.9 (2.5) | 25.1 (5.0) | 12.1 (7.6) |
| Viscera | 0.9 | 18.9 | 2.9 | 6.7 (2.0) | 16.9 (2.9) | 36.6 (5.6) | 17.1 (9.4) |
| Roe (phosphatide) | 1.3 | 25.0 | 0.9 | 2.1 (2.0) | 20.2 (2.7) | 34.4 (7.1) | 16.1 (10.0) |
| Roe (glyceride) | — | 20.4 | 2.0 | 7.0 (2.0) | 30.8 (3.1) | 28.7 (7.3) | 11.1 (7.3) |

At least 96.5 per cent. of the unsaturated C_{18} acids consisted of octadecenoic acid together with traces of octadecatetraenoic acids; there was no indication of the presence of either linolic or linolenic acid.

F. A. R.

Alkaloid-content of *Extractum Hyoscyami*. J. A. C. Van Pinxteren. (*Pharm. Weekblad*, 1939, 76, 1629–1631.)—The 2nd Supplement of the 5th Edition of the Dutch Pharmacopoeia is criticised in respect of its specification for *Extractum Hyoscyami*. It is stated that the alkaloid-content should be 0.125 to 0.150 per cent., although the only requirement of the material from which the extract is made is that it must be of Dutch origin. The method of preparation suggests that it is assumed that the alkaloid-content of the leaves used should be approximately 0.030 per cent. Data obtained by Van der Willen (*Commentaar, Ned. Pharm.* II. 487) vary between 0.02 and 0.1 per cent., whilst those found by Goddijn

(*Pharm. Weekblad*, 1927, 81, 118) for the leaves of *Hyoscyamus niger* were 0.040 to 0.050 per cent. Extracts prepared by the author between April, 1933, and November, 1939, had alkaloid-contents of 0.20 to 0.65 per cent. (11 extracts), and the figure specified by the German Pharmacopoeia is 0.47 to 0.55 per cent. The prescribed maximum doses of *Extractum Hyoscyami* and *Extractum Belladonnae* are 300 and 80 mg. per day, respectively, the specified alkaloid-content for the latter being 1.3 per cent. It is apparent that the prescribed alkaloid-contents and maximum doses for these two products are not in the same ratio. The addition of sugar to such preparations is permitted in order to reduce the alkaloid-content to the required figure when this is exceeded. This, however, may involve difficulties in practice, as the quantity of sugar required may be very large (e.g. 4 times the weight of the *Extractum*), so that it may then more correctly be described by the term "*Syrupus*." J. G.

Detection of Novocaine and Identification of Pantocaine. F. Bledbach and H. Wiegand. (*Scient. Pharm.*, 1939, 9; *J. Pharm. Belg.* 1939, 21, 1025.)—Ten mg. of the substance are dissolved in 30 drops of water and treated with 1 drop of 1:10 sodium nitrite solution, then with several drops of 2.5 per cent. hydrochloric acid, and finally with 20 drops of 2 per cent. phenol solution; an orange colour is produced. With novocaine there form immediately plate-shaped golden-yellow crystals, whilst with larocaine clusters of needles form after about 10 minutes. Addition of sodium hydroxide causes a red colour. The novocaine compound decomposes at about 150° C. and that from larocaine at about 120° C. When a crystal of various anaesthetics in 1 drop of water is treated with 1 drop of 10 per cent. nitrite solution pantocaine alone gives clusters of colourless needles, probably consisting of the nitrite of the base. E. M. P.

Precipitation of Alkaloids with Cuprous Chloride. J. J. L. Zwicker and A. Krusysse. (*Pharm. Weekblad*, 1940, 77, 18–22.)—It has been shown (ANALYST, 1934, 59, 833) that cuprous chloride produces an insoluble precipitate with cardiazol, and the reaction has now been applied as a general test for alkaloids. The reagent is best prepared as follows:—To a solution of 200 mg. of cupric chloride in 1 ml. of water in a graduated test-tube is added a solution of 250 mg. of crystalline sodium sulphite in 5 ml. of water. A white precipitate of cuprous chloride is produced immediately, and on addition of 2 ml. of 4 N hydrochloric acid and sufficient water to bring the volume to 10 ml., this dissolves rapidly. Cuprous bromide (prepared by the precipitation of copper sulphate with potassium bromide and reduction with sulphur dioxide) dissolved in hydrobromic acid may also be used, but is less sensitive. As a rule one volume of the reagent should be added to 4 volumes of a solution of a salt of the alkaloid, but with the cinchona alkaloids it is necessary to reverse these proportions and to use a 5 per cent. solution of the sulphate of the alkaloid in question. Under these conditions quinidine and cinchonidine in particular, produce a precipitate which is amorphous at first, and then deposits thick yellow crystals; 4 volumes of a 1:500 solution of the former will produce with 1 volume of the reagent a turbidity which is, however, redissolved if the volume of reagent is more than doubled. After 2 hours the precipitate produced with quinine is still amorphous, whilst that from cinchonine

has disappeared. This instability, which is characteristic of these precipitates in general, is attributed to re-oxidation of the cuprous ion by the air, and it is therefore particularly marked when the reaction is carried out as a drop-test on a microscope slide. The hydrochlorides of the following alkaloids are soluble and give a negative reaction:—atropine, colchicine, coniine, cytisine, ephedrine, homatropine, morphine, nicotine, novocaine, physostigmine, pilocarpine, piperine, scopolamine, solanine, sparteine and tropine; non-alkaloids which behave similarly are acetanilide, adrenaline, antipyrine, pyramidon, tyrosine, urea and urethane. Positive reactions are obtained with the hydrochlorides of aconitine, apomorphine, berberine, brucine, cevadine, cinchonidine, cinchonine, cocaine, codeine, caffeine, cotarnine, dionine, emetine, heroine, hydrastine, quinidine, quinine, narceine, narcotine, papaverine, strychnine, thebaine, theophylline (the hydrochloride of theobromine is insoluble), veratrine and yohimbine; urotropine behaves similarly, the sensitiveness of the reaction in this instance being 1 : 500,000, although for the above alkaloids it is usually about 1 : 1000. As a rule the crystals are needle-shaped, but papaverine produces square plates, often in rosette-shaped groups (sensitiveness 1 : 2000) and similar to those formed by papaverine and mercuric chloride. Hydrastinine is exceptional in that it forms needle-shaped prisms which are strongly doubly-refracting and appear deep yellow between parallel and crossed nicols. On rotating one of the nicols, however, there is no change in the appearance of the field, and no interference colours are apparent; monochromatic (sodium) light and white light give similar results. When, however, the reagent is prepared by decanting the liquid from the precipitate of cuprous chloride while it is cooling, and dissolving the residual precipitate in 2 ml. of 4 *N* hydrochloric acid, needle-shaped crystals which behave normally when observed between crossed nicols, are obtained 5 minutes after the addition of a little solid hydrastinine hydrochloride to a drop of the reagent on a microscope slide. It is to be noted that whilst morphine does not react, codeine, dionine and heroine do, and this suggests that the presence of a phenolic hydroxy-group may influence the reactivity. Cocaine reacts, whereas tropine, atropine and scopolamine (as hydrochlorides) do not, unless the concentration is at least 2 per cent., when bundles of fine needles are slowly formed; these, however, are rapidly oxidised. The difference in behaviour between cocaine and novocaine is useful for the examination of anaesthetics, especially if oxidation of the compound formed by the former is inhibited by carrying out the reaction in a test-tube under a layer of benzene.

J. G.

Determination of Iodine in Thyroid and its Preparations by Cerate Oxidimetry. W. W. Hilty and D. T. Wilson. (*Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 637–639.)—The method of Lewis (*Ind. Eng. Chem., Anal. Ed.*, 1936, 8, 199) for the determination of iodide by means of ceric sulphate and that of Smith ("*Ceric Sulphate*," Vol. I, 3rd Ed., 1935: "*Ortho-phenanthroline*," 1935, G. F. Smith Chemical Co., Columbus, Ohio), depending upon volumetric oxidation with ceric sulphate and the application of *o*-phenanthroline as indicator have been applied to the determination of iodine in thyroid preparations. When thyroid gland preparations are fused with sodium carbonate, both organic and inorganic

iodine combine with sodium to form sodium iodide. This is extracted with water, and the resulting solution is acidified with hydrochloric acid and titrated with a standard ceric sulphate solution (0.005 *N*) by which the iodide is quantitatively oxidised to iodate. In the determination and in the standardisation of the ceric sulphate solution against ferrous sulphate solution the indicator consists of a solution of 1.485 g. of *o*-phenanthroline monohydrate in 100 ml. of 0.025 *M* ferrous sulphate solution. The procedure is as follows:—Finely powdered thyroid gland (1 g.) is mixed in a nickel crucible with 15 g. and covered with 10 g. of anhydrous sodium carbonate. The crucible is heated first over a burner at dull red heat, and then for 30 minutes in a muffle furnace at a temperature not exceeding 500° C. The fused mass is extracted with warm water and filtered, and the carbonaceous residue is washed until free from soluble salts. The combined filtrate and washings are neutralised to litmus paper with conc. hydrochloric acid and acidified with 20 ml. of the acid per 100 ml. After addition of the indicator (1 drop per 150 ml.) the solution is titrated with 0.005 *N* ceric sulphate solution to a bluish-green end-point persisting for one minute. One ml. of ceric sulphate solution \equiv 0.0003178 g. of iodine. Attempts to incorporate an oxidising agent in the fusion mixture led to inconsistent results, and the use of sulphuric or nitric acid to dissolve the fused mass caused high results. The accuracy of the method was tested by determining known amounts of potassium iodide in admixture with lactose. The error varied from +0.06 to -0.22 on an iodine-content of 60 per cent. The corresponding error in the U.S.P. method of assay applied to the same material varied from +1.39 to +1.59. For the determination of iodine in thyroid tablet preparations, the amount of sample taken should correspond with at least 259.2 mg. (4 grains) of thyroid gland. For calculation it may be assumed that thyroid gland contains 0.200 per cent. of iodine. The method may be modified to make it applicable to the determination of thyroxine in thyroid gland and its preparations.

A. O. J.

Volumetric Estimation of Lac in Glazed Candies. N. M. Molnar and J. Grumer. (*Ind. Eng. Chem., Anal. Ed.*, 1939, **11**, 673-674.)—The Federal Food, Drug and Cosmetic Act (75th Congress, S-5, Sec. 402D) allows the use of lac free from arsenic or lead as a coating on candies, but not in excess of 0.4 per cent. The proposed method for its determination depends upon the solubility of the lac acids in alkali (Weinberger and Gardner, *Ind. Eng. Chem.*, 1938, **30**, 454), and for convenience in the preparation of standard solutions sodium carbonate was chosen as the alkali. The reaction is complete when the solution contains only the sodium salts of the lac acids and sodium bicarbonate (Murty, *Ind. Eng. Chem.*, 1939, **31**, 239), and phenol red was found to be the most suitable indicator. Rogers ("*Manual of Industrial Chemistry*," 5th Ed., p. 1103, New York, D. Van Nostrand & Co., 1931) gives 8, and Murty (*loc. cit.*) 7.7, as the equivalent value of lac in terms of sodium carbonate. Experiments with pure lac confirmed the figure 8. The glazed candy (50 g.) is covered with alcohol 3A (5 parts of methyl alcohol added to 100 parts by volume of alcohol) or 2B (0.5 part of benzene added to 100 parts by volume of alcohol). As a rule, less than 40 ml. of alcohol suffice, and the mixture is allowed to stand, with occasional stirring, for an hour. The

alcoholic solution is decanted through a filter, and the candy is re-extracted with 10 ml. of alcohol and finally washed with sufficient alcohol to provide 50 or 100 ml. of solution. An aliquot portion (20 ml.) of the filtrate is mixed with 20 ml. of water, and the mixture is kept at boiling point for at least five minutes. After the addition of 3 to 5 drops of phenol red (0.1 g. dissolved in 28.2 ml. of 0.01 *N* sodium hydroxide solution and diluted to 250 ml.), the liquid is titrated while hot with standard sodium carbonate solution containing 1 mg. of sodium carbonate in each ml. The titration is continued until the red colour matches that formed when 1 or 2 drops of the sodium carbonate solution are added to a control amount of alcohol similarly treated. The number of ml. of alkali, multiplied by 8, gives the number of mg. of lac in the aliquot portion taken. Organic acids, particularly citric acid, may be present in some candy. When their presence is suspected, or when the content of lac appears high, it is advisable to titrate an aqueous extract of the candy and deduct the result from the result of the alcoholic titration. If fatty acids are present, they are extracted by means of carbon tetrachloride and, after removal of the solvent, dissolved in alcohol and titrated with the sodium carbonate solution. Since lac is soluble in carbon tetrachloride to the extent of 6.8 per cent., the figure found requires a corresponding correction. The lac remaining in the candy is then determined by the process described. Experiments with candy to which known amounts of lac had been added showed that the method is accurate.

A. O. J.

Biochemical

Determination of Magnesium in Biological Material. J. P. Nielsen. (*Ind. Eng. Chem., Anal. Ed.*, 1939, **11**, 649-651.)—The principal objection to methods for the determination of magnesium by means of 8-hydroxyquinoline (Cruess-Callighan, *Biochem. J.*, 1935, **29**, 1081; Greenberg, Anderson and Tufts, *J. Biol. Chem.*, 1935, **111**, 561; Greenberg and Mackey, *ibid.*, 1932, **96**, 419; *Abst., ANALYST*, 1932, **57**, 730) is probably in connection with the bromination of the 8-hydroxyquinoline. Greenberg, Anderson and Tufts (*loc. cit.*) have suggested a procedure involving the use of special apparatus to prevent loss of bromine. Since magnesium quinolate can be precipitated with constant composition, any measurable reaction involving the organic part of the molecule might be used in the determination of the precipitated magnesium, and experiments showed that this can be effected by the use of ammonium hexanitrate cerate as an oxidising agent, thus eliminating the troublesome bromination necessary in the previous methods. The sample of biological material, containing from 0.05 to 3.00 mg. of magnesium (as magnesium oxide), is treated with sulphuric acid, evaporated to dryness, and ignited in a muffle furnace at 500° to 600° C. The ash is dissolved in a few ml. of 6 *N* nitric acid and evaporated to dryness (this stage may be omitted if the product does not contain tin). The residue is warmed with 1 ml. of conc. hydrochloric acid and diluted with several volumes of water, and the tin oxide and silica are removed by filtration. Traces of tin may dissolve again in the hydrochloric acid, but will be removed with the iron and aluminium later. A few drops of bromine are added, the excess bromine is removed by boiling, and

the pH is adjusted to about 4 with dilute sodium hydroxide solution, bromocresol green being used as indicator; the final adjustment is made with 20 per cent. sodium acetate solution. A slight excess of phosphate is necessary to precipitate iron at this pH value. After removal of the iron precipitate, the filtrate is treated with 1 ml. of saturated sodium oxalate solution, the pH is adjusted to 4.4 or 4.6 with dilute oxalic acid solution, and the liquid is boiled, cooled and allowed to stand for 2 or 3 hours. The precipitate of calcium oxalate is filtered off and washed with ammonia water (1 : 50). The filtrate is acidified to the yellow shade of the indicator with dilute hydrochloric acid, concentrated to about 10 ml. and treated with 5 ml. of 8-hydroxyquinoline reagent (1 g. of 8-hydroxyquinoline in a solution containing 89 ml. of absolute alcohol, 10 ml. of conc. ammonium hydroxide and 1 ml. of conc. hydrochloric acid). The mixture is maintained at a temperature just below its boiling-point for 15 minutes. When cold the supernatant liquid is removed by means of a sintered glass filter-stick having a thin layer of asbestos over the disc, and the beaker is washed once with 95 per cent. alcohol and six to eight times with *N* ammonia until all the alcohol has been removed. The precipitate is dissolved in 5 ml. of 2 *M* perchloric acid, and the solution is diluted to such a volume that 5 ml. contains from 0.05 to 0.10 mg. of magnesium oxide. This aliquot portion is treated with 5 ml. of 0.05 *N* cerate reagent and heated in a water-bath at 96° to 100° C. (The cerate reagent is prepared by dissolving 14.5 g. of ammonium hexanitrate cerate, $(\text{NH}_4)_2\text{Ce}(\text{NO}_3)_6$, in 500 ml. of 2 *M* perchloric acid and heating the solution in a boiling water-bath for two hours. After an interval of two days the solution is standardised against 0.02 *N* ferrous ammonium sulphate solution.) The excess of cerate reagent in the solution is titrated with standard ferrous ammonium sulphate solution, *o*-phenanthroline ferrous sulphate solution being used as indicator. One mole of magnesium corresponds with 59.7 equivalents of cerate reagent. As a representative application of the method the magnesium-content of the ash of tomatoes was determined, and the results agreed well with those found by the official method of the A.O.A.C.

A. O. J.

Determination of Small Quantities of Molybdenum in Biological Materials. D. Bertrand. (*Bull. Soc. Chim.*, 1939, 6, 1676-1689.)—For the determination of a few γ of molybdenum in 100 g. sample of dry material the following process was found satisfactory. *Destruction of organic matter.*—Calcination is preferred to wet oxidation. G. Bertrand's two-stage process is advantageous. The sample is carbonised below 500° C., the carbonaceous mass is extracted with hot water to remove salts, the residue is ashed, and the aqueous extract is evaporated to dryness along with the ash. The residue is fused with sodium carbonate in a platinum vessel; the melt is decomposed with hydrochloric acid, and the liquid is evaporated to dryness to render silica insoluble. The residue is dissolved as far as possible in hydrochloric acid, and the silica is filtered off. (Spectroscopic tests on the silica obtained indicated freedom from molybdenum.) *Separation of molybdenum.*—It is necessary to isolate the molybdenum as far as possible from various salts, such as phosphates, present in the solution of the ash, which interfere with the final sulphide colorimetric determination of molybdenum. A cupferron-extraction method is suitable. The acid solution, obtained as described above,

is diluted so that its salt concentration is below 5 per cent.; the pH is adjusted to about 1.6 (change point of thymolsulphonephthalein indicator), and the solution (100 to 150 ml.) is extracted several times with 10-ml. portions of chloroform after the addition of successive portions of a freshly-prepared 10 per cent. solution of cupferron. The combined chloroform extract, which contains the molybdenum together with copper, iron and traces of tin, is evaporated to dryness. Cupferron in the residue is destroyed by heating with sulphuric and nitric acids, and the bulk of the acids used is subsequently removed by evaporation. The residue is dissolved in 5 to 6 ml. of water with the addition of 0.3 ml. of conc. hydrochloric acid. Iron and tin are precipitated by adding a little hydrogen peroxide and rendering the liquid slightly ammoniacal; the precipitate is separated by centrifuging. Re-precipitation is advised. The precipitate is rejected. The solution is evaporated to dryness, and the residue is dissolved in 3 ml. of $N/10$ hydrochloric acid. *Determination of molybdenum.*—The solution is treated with 1 ml. of 10 per cent. sodium sulphide solution and heated to 70°C ., and hydrogen sulphide is passed into it for a few seconds. The precipitate (copper sulphide) is filtered off and washed with a little water. To the filtrate, diluted to 9 ml., is added 1 ml. of $N/5$ ammonium chloride solution, and the liquid is boiled for 10 to 20 seconds and cooled. The colloidal solution of molybdenum sulphide thus produced is reasonably stable, varying little in colour in 15 to 20 minutes, and thus allowing the molybdenum to be determined colorimetrically. A photo-electric colorimeter was used by the author; the sensitiveness—1 to 2γ of molybdenum in 10 ml. (0.0002 g. per litre)—was somewhat greater than could be obtained by visual comparison with standards. It was concluded that the lower limit determinable was 0.002 mg. of molybdenum in 100 g. of dry organic matter; the accuracy is estimated to be about 2 to 3 per cent. when 0.05 mg. or more of molybdenum in 100 g. is present.

S. G. C.

Rôle of Sorbitol in the Carbon-Metabolism of the Kelsey Plum.

I. Changes in Chemical Composition during Growth and Storage. I. Donen. (*Biochem. J.*, 1939, **33**, 1611–1620.)—In extracts of the Kelsey plum, prepared by extracting the minced fruits (250 g.) with 75 per cent. alcohol, and evaporating off the alcohol to obtain an aqueous solution (250 ml.), sorbitol was identified by the following method. A portion (5 ml.) of this solution was fermented to remove sugars and then treated with 0.5 ml. of benzaldehyde and 1 ml. of 50 per cent. sulphuric acid; dibenzal-sorbitol of m.p. 172° to 174°C . was precipitated. This derivative was also employed to estimate the amount of sorbitol present, according to the method of Martin* (*Bot. Gaz.*, 1937, **99**, 48), the accuracy of which was

* In this method 75 ml. of the extract are pipetted into a flask and 1 g. of activated charcoal is added. The flask is tightly corked and well-shaken, and the solution is immediately filtered into a dry flask. A 50-ml. aliquot portion of the filtrate is pipetted into a beaker and evaporated on a steam-bath to a thick syrup. One ml. of 50 per cent. sulphuric acid and 0.5 ml. of benzaldehyde are added, and the mixture is stirred for 5 minutes and then placed in the refrigerator for about 20 hours. To the thick yellowish paste 50 ml. of cold distilled water are added, and the paste is broken up with a stirring rod, and left in the refrigerator for a further two hours. A slight excess (1.5 g.) of anhydrous sodium carbonate is added, and finally, 1 hour later, the material is removed from the refrigerator, boiled for 1 to 2 minutes, and cooled in running water. The white, amorphous precipitate of dibenzal-sorbitol is collected on a tared Gooch crucible, washed thoroughly with water, dried for 10 to 12 hours at 100°C . and weighed. One hundred mg. of sorbitol are equivalent theoretically to 186.8 mg. of dibenzal-sorbitol. The method gives almost quantitative yields over a range of 60 to 250 mg. of sorbitol.

confirmed by tests on pure sorbitol. Reducing sugars, sucrose and fructose were determined by the method of van der Plank (*Biochem. J.*, 1936, 30, 457). In addition the dry weight, the soluble (in both 75 per cent. alcohol and in water) solids and the acidity of the plum extracts were estimated. The complete series of analyses was carried out on samples of plums picked (usually from the same trees) at regular intervals throughout the season, and again on similar samples after varying periods of storage, in order to determine the general effect of maturity on loss of sorbitol and sugar in store. It was found that on the average the mature Kelsey plum contained 16.65 g. of dry solids per 100 g. of fresh weight, this being made up of 1.5 g. of alcohol-insoluble residue (mainly hemicelluloses and pectins), 10.48 g. of sugar, 2.82 g. of sorbitol, 0.835 g. of acid calculated as malic acid, 0.35 g. of soluble organic nitrogenous material and 0.4 g. of ash. Sorbitol appeared to be stored only during the later part of the growth cycle, and in place of hexoses, when the latter had reached a maximum concentration. Loss of respirable material from stored plums occurred at the expense of sorbitol, sugar and acid. Sorbitol was rapidly lost, but the loss of sugar depended on the initial sorbitol concentration, and at 13° C. and 25° C., mature plums of high sorbitol-content showed no significant loss of sugar. Plums of low sorbitol-content showed marked loss of sugar only when most of the sorbitol had disappeared. When plums were stored at 1° C. for 25 days and then at 7.5° C. or 20° C., they showed a 10 to 15 per cent. increase in total sugar, and a slightly greater decrease in the amount of sorbitol.

F. A. R.

Vitamin A and Carotenoids in the Liver of Mammals, Birds, Reptiles and Man. H. B. Jensen and T. K. With. (*Biochem. J.*, 1939, 33, 1771–1786.)—The amounts of vitamin A and carotenoids in the livers of 21 species of mammals, 36 species of birds, 2 species of reptiles and in 8 human specimens were estimated. The livers were saponified and extracted with ether, and the extracts were used for (a) the measurement (in alcoholic solution) of the absorption spectrum, (b) the measurement of the Carr–Price reaction by means of a Zeiss–Pulfrich photometer using three different filters, (c) the colorimetric estimation of carotenoids, and (d) micro-chromatographic analysis to determine the nature of the carotenoids present. Most of the livers gave absorption spectra typical of vitamin A. It was found that the relation between the extinction value of the solution obtained in the Carr–Price reaction using filter S61, and $E_{1\text{ cm}}^{1\%}$ ($328m\mu$) had a fairly constant value of 2.60 to 2.75. Considerable variation was encountered in the amounts of vitamin A and carotene present in the livers even of closely related species or of species that exist on similar kinds of food. There appeared to be no simple relationship between the amount of vitamin A and carotene in the food ingested and their concentrations in the liver. Carotene was found to be present in considerable amounts in the livers of certain beasts of prey (*e.g.* foxes) and birds as well as in herbivora, but most of the livers examined were practically free from carotenoids.

F. A. R.

Thiochrome Test for Aneurin in Urine as an Index of Nutritional Level. G. M. Hills. (*Biochem. J.*, 1939, 33, 1966–1979.)—The samples of urine were preserved at an acid pH, 0.5 ml. of conc. hydrochloric acid or glacial acetic

acid being added; toluene was added as well to prevent mould growth. After filtration, to remove any deposit, the urine was diluted so that the amount excreted per hour was made up to 500 ml. The pH was then adjusted to 3-5, and two 75-ml. portions of the urine were each stirred mechanically in a centrifuge tube for 1 to 2 minutes with 25 ± 2 mg. of "Clarit" acid clay. Other aliquot portions (usually two) were similarly treated after the addition of different amounts of aneurin (less than 4 γ). The adsorbates were collected by centrifuging for 2 to 5 minutes, and the supernatant liquors were discarded. The wet adsorbates were treated in the same tubes with 2 ml. of pure methyl alcohol. To provide a blank, 1 ml. of water was added to one of the tubes not containing added aneurin. The contents of the four tubes were then stirred with a stream of nitrogen, and 1 ml. of 7.5 *N* sodium hydroxide solution, followed by 1 ml. of 1.25 per cent. potassium ferricyanide solution, was added to each of the tubes except the blank. Isobutyl alcohol (12.5 ml.) saturated with water was then added to all four tubes, and the contents were stirred for 1 to 2 minutes. After standing for a few minutes the upper layer was decanted through a dry 8.5-cm. filter-paper (previously purified by exhaustive extraction with wet isobutyl alcohol) into a test-tube specially selected for fluorescence measurement and calibrated at 10 ml. When 10 ml. of the solution had been collected, the tube was stoppered and the fluorescence was measured in an improved type of Cohen fluorometer. The fluorescence (expressed as a percentage of that produced under similar conditions from 1 γ of aneurin, oxidised in pure solution with 0.3 ml. of 1.25 per cent. potassium ferricyanide solution) was plotted against the amount of added aneurin, and the curve so obtained was produced backwards until it cut the abscissa drawn at a level corresponding to the fluorescence of the unoxidised blank. The length of the intercept so produced gave the aneurin-content of the sample. The most important modification introduced into the fluorometer consisted in the use of a blue colour-filter (Wratten 49A or 48 with Weston "Photronic" cells types I or II, respectively) to bring the maximum response of the cell to a wave-length of 460 to 470 $m\mu$, corresponding with the maximum fluorescence of thiochrome.

F. A. R.

Distribution of Aneurin in Foods determined by Chemical Analysis.

M. Pyke. (*J. Soc. Chem. Ind.*, 1939, 58, 338-340.)—The thiochrome method of assaying aneurin has been applied to foodstuffs, with results that agree well (with certain exceptions) with the values obtained by the rat bradycardia method. A novel method of extracting the vitamin has been used. To 20 g. of the finely-minced or powdered foodstuff is added a solution of 0.1 per cent. of pepsin in 0.33 per cent. hydrochloric acid, and the volume is made up to 97.4 ml. The mixture is incubated overnight at 37° C., 2.6 ml. of *N* sodium hydroxide solution and 100 mg. of takadiastase are added, and incubation is continued for a further 5 hours. In this way the aneurin released from the tissues of the foodstuff and its pyrophosphate is hydrolysed to the free vitamin. A part of the solution is centrifuged (not filtered) and two 3-ml. portions are pipetted into graduated cylinders containing the following reagents kept stirred by means of a stream of nitrogen. The first cylinder contains 2 ml. of methyl alcohol, 1 ml. of 30 per cent. sodium hydroxide

solution and 1 ml. of 1 per cent. potassium ferricyanide solution. The second cylinder contains 2 ml. of methyl alcohol and 1 ml. of sodium hydroxide solution only. After 1 minute the solutions are made up to 10 ml. with water, 13 ml. of isobutyl alcohol are added, and the solutions are well mixed. Ten ml. of the supernatant layer in each tube are pipetted into uniform test-tubes, and 1 ml. of alcohol is added to each. A standard solution of thiochrome (in isobutyl alcohol) is added, 0.1 ml. at a time, to the aliquot portion prepared without ferricyanide, until the intensities of fluorescence match when the two tubes are held side by side at an angle of 45° against a nickel oxide filter arranged in a vertical position to transmit the ultra-violet light of a mercury vapour lamp. The standard solution of thiochrome is prepared by treating a solution containing a known amount of aneurin in the same way as in the test. The following represents some of the more important results obtained, values being expressed in I.U. per 100 g.—apples (different varieties) 6 to 23; apricot, fresh 23, dried 57; bacon, raw 88; banana 8; beans, tinned-baked 17; raw butter 140; beef 23; biscuits 0 to 19; bread, wholemeal 50, proprietary brown 50 to 95, white 13; cabbage 23; carrot, raw 15; cauliflower 49; cheese (18 different kinds) 0 to 21; chocolate 10; cod's roe, smoked 300; egg 86; hake 13; halibut 38; ham, cooked 190; herring 2; herring roe (hard) 10; honey 6; jam (apricot, blackcurrant, raspberry, strawberry) 0, (greengage, plum) 25; kidney, ox 57, sheep 76; lentils 170; lettuce 23; liver, calf 66, ox 61; milk, fresh 10, condensed sweetened 32, dried skimmed 85; mutton 34; oatmeal, breakfast 95; peanut 228; peas (green) 120; plaice 65; pork 475; potatoes, boiled or fried 40; salmon, tinned 38; sugar 2; tomato 15; veal 57.

F. A. R.

Estimation of Vitamin B₁ in Cerebro-spinal Fluid. H. M. Sinclair. (*Biochem. J.*, 1939, **33**, 1816–1821.)—The method previously described (*cf.* ANALYST, 1939, **64**, 214) has been applied to cerebro-spinal fluid. From 1 to 4 ml. of the latter are added to flasks containing the medium (with 0.4 per cent. of asparagine), the flasks are sterilised by tyndallisation and inoculated with *Phycomyces blakesleeanus*. The weight of the fungus produced after 10 days is proportional to the amount of aneurin present. The presence of the vitamin is confirmed by the fact that no growth is obtained when the vitamin is converted into thiochrome by alkaline potassium ferricyanide solution. Cerebrospinal fluid, like blood, contains an adjuvant factor, but in much smaller amounts. Consequently the values obtained for the aneurin-content are higher than the true value, but a correction can be applied for this source of error by multiplying the weight of fungus obtained with 1 ml. of c.s.f. (or blood) by the factor:

$$\frac{\text{weight obtained with excess vitamin}}{\text{weight obtained with 1 ml. of c.s.f. (or blood) in presence of excess vitamin}}$$

The method estimates aneurin both in the free state and combined as cocarboxylase; normal c.s.f. contains none of the latter. The method was applied to 272 samples of c.s.f. obtained from hospital patients, but apart from the fact that pathological samples tended to give higher values than normal samples, the method appears to be of no clinical value.

F. A. R.

Estimation of Aneurin in Blood. II. Further Modification of Melklejohn's Method. H. M. Sinclair. (*Biochem. J.*, 1939, 33, 2027-2036.)—The method of assaying aneurin by measuring the growth-rate of *Phycomyces blakesleeanus* (cf. ANALYST, 1939, 64, 214, and preceding abstract) does not give satisfactory results for blood, since factors are present in blood that have an adjuvant action. Thus samples of 1, 2 and 3 ml. of blood do not usually give the same values per unit volume. The same values can, however, be obtained by multiplying the weight of fungus obtained in the presence of blood by the factor:

$$\frac{\text{weight of fungus obtained with excess aneurin}}{\text{weight of fungus obtained with excess aneurin and blood}}$$

Four 2-ml. samples of the blood, two without and two with added excess aneurin (2.5γ), are incubated with the organism as previously described, and the mycelia are filtered off, dried and weighed. The amount of growth produced by a solution of 2.5γ of aneurin is ascertained in a similar way. The weight produced by the blood with the excess aneurin is then multiplied by the factor given above and the corrected value is converted into amounts of aneurin from the growth-vitamin curve obtained with the pure substance.

F. A. R.

Quantitative Estimation of Nicotinic Acid in Urine. E. Bandier. (*Biochem. J.*, 1939, 33, 1787-1793.)—The colour reaction previously described (Bandier and Hald, cf. ANALYST, 1939, 64, 441) has been applied to the estimation of nicotinic acid in urine. Five ml. of urine and 2 g. of sodium chloride are thoroughly mixed in a 50-ml. graduated flask fitted with a glass stopper, and 45 ml. of acetone are added from a burette. The mixture is shaken to extract the nicotinic acid and then centrifuged. Twenty ml. of the acetone extract (equivalent to 2 ml. of urine) are pipetted off and transferred to a flask, together with 3 ml. of water. The acetone is evaporated, and the residual solution is transferred quantitatively to a 20-ml. graduated flask with the aid of 5 ml. of 2 per cent. potassium dihydrogen phosphate solution. The flask is heated for 5 minutes on a water-bath at 75° to 80° C., 1 ml. of a freshly-prepared 4 per cent. solution of cyanogenbromide is introduced, and, after 5 minutes' further heating, the flask is cooled. Ten ml. of a freshly-prepared saturated solution (5 per cent.) of metol are added, and the solution is diluted to 20 ml. After standing for 1 hour at room temperature with protection from the light, the colour of the solution is measured in a Pulfrich photometer (filter S43) against a solution containing the same amounts of reagents made up to 20 ml. with distilled water. The blank is prepared by treating a second 20-ml. portion of the acetone extract as described above and adding to it 0.18 ml. of 2 N sulphuric acid instead of metol. The colour is measured in the Pulfrich photometer (filter S43) with distilled water in the other cell. The value found, corrected, of course, for the thickness of the cell, is subtracted from the first reading, the nicotinic acid content being calculated from the difference by comparison with readings obtained with a standard solution. In some instances the blank value may amount to 6 times the value of the colour reaction proper.

The reaction is given, not only by nicotinic acid, but also by nicotinamide (free and combined as co-enzyme), nicotinuric acid and nicotine, the relative

colours produced by equimolar concentrations of these 4 substances being, respectively, 100, 142, 42, 8. Trigonelline and methylpyridinium hydroxide do not react. The above-mentioned derivatives of nicotinic acid are converted into the acid itself by alkaline hydrolysis, a procedure that enables a fairly accurate estimation of the total (free and combined) nicotinic acid to be made. Fifteen ml. of urine and 2 ml. of 10 *N* sodium hydroxide solution are mixed in a 20 ml. graduated flask which is closed with non-absorbent cotton wool and then heated for 30 minutes on a boiling water-bath. After cooling, the *pH* of the solution is adjusted to 5 by the addition of conc. hydrochloric acid (about 1.6 ml.), and the volume is made up to 20 ml. After centrifuging, the analysis is continued as described above.

The amount of nicotinic acid found after hydrolysis was 2 to 3 times that found prior to hydrolysis. Recoveries ranging from 96 to 103 per cent. (10 experiments) of added nicotinic acid were obtained. After the oral ingestion of 90 mg. of nicotinic acid, 14 per cent. thereof was found to have been excreted, chiefly during the first hour. Some of this was in the form of nicotinuric acid or a closely related substance.

F. A. R.

Estimation of Nicotinic Acid in Urine. L. J. Harris and W. D. Raymond. (*Biochem. J.*, 1939, 33, 2037-2051.)—A careful study of König's reaction was made, and as a result *p*-aminoacetophenone was selected for use in the test in preference to other aromatic amines. Furthermore, since the coloured substances formed in the reaction were found to be sensitive to light and to changes in *pH*, modifications were introduced to overcome the discrepancies arising from these causes. To a 25-ml. specimen of the urine, 5 ml. of 20 per cent. sodium hydroxide solution are added and the mixture is heated for 30 min. on a steam bath to convert any nicotinamide into free nicotinic acid. The solution is then treated with 2 ml. of 4 per cent. sodium bicarbonate solution (to help stabilise the end-point) and neutralised accurately to *pH* 6 by the cautious addition of conc. hydrochloric acid from a micro-burette (bromothymol blue as external indicator). The contents of the flask are transferred to a 50-ml. graduated flask and made up to the mark. Four 10-ml. portions of the prepared urine are transferred to four 15-ml. standard flasks, into two of which 0.2 ml. and 0.4 ml. of a standard solution of nicotinic acid (100 γ per ml.) have previously been introduced. All the flasks are then immersed for about 10 min. in a bath kept at 80° C. and shaded from the light. Two ml. of freshly-prepared cyanogen bromide solution (a 10 per cent. solution of potassium cyanide is added, drop by drop, to a saturated solution of bromine until decolorised) are added to each of the flasks except one of those not containing added nicotinic acid, which is to serve as blank. After being rotated to mix the solutions, the flasks are allowed to remain for a further 4 minutes at 80° C., and are then immersed in a bath of cold water for 4 minutes. To each is then added 0.2 ml. of amine solution (made by dissolving 5 g. of *p*-aminoacetophenone in 14 ml. of 10 per cent. hydrochloric acid and diluting to 50 ml.), the contents of the flasks are mixed, and the flasks are placed in the dark for 15 minutes. To each is then added 0.4 ml. of 10 per cent. hydrochloric acid and, after standing for a further 15 minutes in the dark, the solutions are introduced in succession into the 3-cm. cell of a Pulfrich photometer and the colours measured

with filter S47. The extinction values thus obtained are plotted against the amount of nicotinic acid added, and the resulting graph (which should be a straight line) is produced backwards until it cuts the abscissa. The distance of the point of intersection from the origin indicates the amount of nicotinic acid in the sample. Good agreements have been obtained in duplicate experiments, with an experimental error within ± 10 per cent., whilst added nicotinic acid or nicotinamide is quantitatively recovered.

F. A. R.

Oxalate Formation in Ascorbic Acid Solutions. A. E. Jurist and W. G. Christiansen. (*Amer. J. Pharm.*, 1939, 111, 347-350.)—Aqueous solutions of ascorbic acid are unstable and progressively deteriorate in strength, while oxalic acid is formed as one of the products of auto-oxidation (Ghosh and Rakshit, *Biochem. Z.*, 1938, 299, 394). The results here described show that oxalic acid is invariably present in solution, the amounts in the samples examined ranging from 0.19 to 11.14 mg. per ml. The amount of oxalic formed is not proportional to the loss of ascorbic acid on ageing. Oxalate was formed in solutions of monoethanolamine ascorbate as well as in those of the sodium and calcium salts. For the determination of the oxalic acid 10 ml. of the solution at pH 5.0 to 7.5 were treated with 2 ml. of 10 per cent. calcium acetate solution in a 15-ml. centrifuge tube, which was then stoppered and left for 5 days. The precipitate was separated and washed with the aid of centrifuging and finally collected on asbestos in a Gooch crucible; as little water as possible was used throughout. Finally the asbestos mat and precipitate were transferred, with 25 ml. of water, to a 150-ml. beaker containing 25 ml. of 20 per cent. sulphuric acid, the mixture was heated to 70° C., and the oxalic acid was titrated with *N*/100 potassium permanganate solution.

Evaluation of the Activity of Powders of *Veratrum viride* by the *Daphnia* Method. I. Cohen. (*Amer. J. Pharm.*, 1939, 111, 426-429.)—When an extract (50 ml.) of *Veratrum viride*, prepared by vigorously shaking 1 g. of the powder with 100 ml. of water (or culture medium) and then filtering, is added to 50 ml. of culture medium (Bovung) containing 50 standardised 10-day old *Daphniae magna*, a change in the behaviour of the organisms is observed. At first, the swimming activity is accelerated and also inco-ordinated, but later the *Daphniae* become exhausted and begin to settle to the bottom of the jar ("debility shift"), where they ultimately lie quiescent. The activity of the *Veratrum* extract can be estimated by noting at intervals how many of the *Daphniae* are swimming above the midmark of the jar and how many lie below it. The jars recommended for this purpose are narrow museum jars 15 × 10 × 2 cm. When appreciable differences are found in the relative activities of two extracts, a more exact comparison can be obtained by diluting the stronger of the two until the behaviour of the *Daphniae* towards both solutions is the same. The results obtained were in agreement with those obtained by assays on rats, rabbits and guinea-pigs.

F. A. R.

Agricultural

Determination of Deguelin in Derris and Cubé. L. D. Goodhue and H. L. Haller. (*Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 640-642.)—The chloroform extract of derris or cubé is easily divided into three fractions. Rotenone can be

removed by crystallisation from carbon tetrachloride (Jones, *J. Amer. Chem. Soc.*, 1931, 53, 2738; Jones and Graham, *J. Assoc. Off. Agr. Chem.*, 1938, 21, 148), and the remaining resin can be divided into alkali-soluble and alkali-insoluble portions (Haller and La Forge, *J. Amer. Chem. Soc.*, 1934, 56, 2415). The last part consists largely of optically active deguelin, since racemic deguelin is obtained by further treatment of this fraction with dilute alkali. Fifty g. of finely ground material containing deguelin were extracted with chloroform in a Soxhlet extractor for 7 hours. After removal of most of the solvent the extract was dissolved in 75 ml. of ether and the solution was extracted with two 15-ml. portions of 5 per cent. potassium hydroxide solution saturated with sodium chloride. The alkaline liquids were extracted with ether which was added to the first ethereal solution and washed once with dilute hydrochloric acid (1 : 10). The alkali-soluble portion was discarded. After removal of the ether, the resin was dissolved in 40 ml. of carbon tetrachloride and the solution, seeded with rotenone carbon tetrachloride solvate, was allowed to crystallise overnight at 0° C. The solvate was then filtered off and washed with ice-cold carbon tetrachloride. The filtrate and washings were evaporated to remove the solvent, the residue was dissolved in 10 to 15 ml. of methanol and placed while warm in a 25-ml. flask with 10 drops of 40 per cent. potassium hydroxide solution. After thorough mixing of the liquids, the flask was filled with warm methanol and closed by means of a cork carrying a funnel made from a drawn-out test-tube in such a manner that no air bubbles remained in the flask and some of the liquid was forced up into the funnel. More methanol was placed in the funnel to counteract contraction on cooling and as a reserve for evaporation, and the solution was maintained at 45° C. for an hour to prevent separation of the resin before it was racemised. Racemisation was not usually complete until the liquid had stood overnight. The racemised deguelin was cooled at 0° C. for an hour. The crystals were separated by filtration without washing, dissolved in chloroform and the chloroform removed by evaporation twice with carbon tetrachloride. Finally, the deguelin was crystallised from 5 or 10 ml. of carbon tetrachloride according to the amount present. The crystals were collected on a tared Gooch crucible, washed with cold carbon tetrachloride saturated with deguelin, dried at room temperature and weighed as the 1:1 deguelin-carbon tetrachloride solvate. The amount of deguelin in the impure solvate was determined by the red colour test (Goodhue, *J. Assoc. Off. Agr. Chem.*, 1936, 19, 118; Abst., *ANALYST*, 1936, 61, 486). It was assumed that deguelin alone was responsible for the colour, and the fact that racemic deguelin gives only 80 per cent. of the colour of rotenone was taken into account when rotenone was used as the standard. Experiments showed that the solubility of deguelin in carbon tetrachloride may be compensated for by adding 0.08 per cent. when 5 ml. of carbon tetrachloride are used and 0.11 per cent. when 10 ml. are used. The amount of deguelin in the samples of derris examined varied from 0.24 to 3.9 per cent. and in the samples of cubé from 0.25 to 2.3 per cent. The high toxicity to insects of the non-crystalline portion of derris and cubé extracts, coupled with a low deguelin content, suggests the presence of unidentified compounds that contribute to the toxicity.

A. O. J.

Organic

Iodimetric Determination of Organic Acids. B. Singh and S. Singh. (*J. Indian Chem. Soc.*, 1939, 16, 343-345.)—Oxalic, tartaric, citric, malic, and glycollic acids can be determined by treatment with an excess of potassium iodide and potassium iodate in presence of a barium, zinc or magnesium salt, followed by potentiometric titration of the liberated iodine against sodium thiosulphate solution. The authors carried out the titration at 10° C., using a platinum electrode coupled with a saturated calomel electrode, and keeping the solution stirred with a mechanical stirrer. A sharp fall in E.M.F. occurs at the equivalence point. E. M. P.

Arylation of Oils and Fats. Synthesis of Tollyl Stearic Acid and its Esters. W. Kimura and J. Tsurugi. (*J. Soc. Chem. Ind. Japan*, 1939, 42, 390-391b.)—Camellia oil (Wijs iodine value, 82.70; acid value, 2.51; ester value, 184.43; saponification value, 186.94) was used as the source of oleic acid (*cf.* Kimura, *J. Soc. Chem. Ind., Japan*, 1929, 32, 459). The optimum conditions for arylation were found to be the presence of a 30 per cent. excess of aluminium chloride, a 1400 per cent. excess of toluene, a reaction-temperature of 25° to 34° C., and a reaction-time of 2 to 3 hours. On fractional distillation *in vacuo* of the resulting methyl esters of the tolylated fatty acids of camellia oil, the unsaturated or unchanged (*i.e.* non-arylated) constituents were obtained as a fraction of low b.p., the reaction-products of high mol. wt. as a fraction of high b.p., or in the residue, and the tolyl stearic acid methyl ester as a pale yellow oil, with b.p. 232° to 245° C. (3 mm.), n_D^{80} , 1.4754 to 1.4781 and sp.gr. (30° C.), 0.9153 to 0.9167. The tolyl stearic acid separated from the ester was a pale yellow viscous oil, the yield of which was lowered by the side-reactions. Constants of the tolylated camellia oil were as follows:—Wijs iodine value, 9.92; acid value, 7.36; ester value, 152.8; saponification value, 160.16; n_D^{80} , 1.4700. This reaction may also be used as a means of separating the saturated and unsaturated constituents of oils and fats, the fatty acid component being any mono-ethylenic acid or halogen fatty acid (or a derivative of these), while the aromatic constituent can be benzene or its homologues or their derivatives (*cf.* Nicolet and De Milt, *J. Amer. Chem. Soc.*, 1927, 49, 1106). J. G.

Occurrence of an Isomer of Ricinoleic Acid in the Fatty Oil from the Seeds of *Vernonia anthelmintica*. N. L. Vidyarthi and M. V. Mallya. (*J. Indian Chem. Soc.*, 1929, 16, 479-480.)—The fatty oil obtained from the seeds of *Vernonia anthelmintica* (N.O. *Compositae*) has been found to have an optical rotation of $[\alpha]_D^{28}$, -10.7 and an acetyl value of 135.1. These values are not entirely due to the high percentage of unsaponifiable matter, since the mixed fatty acids freed from unsaponifiable matter had an optical rotation of $[\alpha]_D^{25}$, -7.2 and an acetyl value of 118.2 and contain about 60 per cent. by weight of a hydroxy straight-chain acid belonging to the monohydroxyoctadecenoic (ricinoleic) series. The acid is viscous and sparingly soluble in petroleum spirit, and has a saponification equivalent of 299.0 (calc. for $C_{18}H_{34}O_2$, 298.2). It differs from ricinoleic acid in being *l*-rotatory, $[\alpha]_D^{28}$, -7.8, and the OH group appears to be in a different

position, for it is readily substituted by halogens even during the period of iodine value determination by the Wijs and Hanus methods, but on acetylation and protection of the OH group no more substitution occurs, and the exact quantity of iodine for the ethylene linkage is absorbed. The iodine value of the acid is 108.4 and that of the acetylated acid 73.8. It does not yield a solid dibromide like that reported for quince oil acid.

D. G. H.

Bamboo Leaf Wax. M. Tsujimoto. (*J. Soc. Chem. Ind. Japan*, 1939, 42, 396B.)—About 1 per cent. of wax was extracted with hot petroleum spirit from the leaves of the bamboo *Sasa paniculata* Makino et Shibata, which grows abundantly in the mountainous districts of Central Japan. When purified by solution in ethyl acetate and treatment with animal charcoal it was a brownish-yellow, hard, brittle solid, having the following properties:—sp.gr. (25°/4° C.), 0.961; m.p., 79° to 80° C.; acid value, 14.5; saponification value, 43.4; iodine value (Wijs), 7.8; unsaponifiable matter, approximately 65 per cent. (a brownish-yellow solid; m.p., approximately 87° to 88° C.; iodine value, 12.7; acetyl value, 58.9; no appreciable quantity of sterols present). The fatty acids were hard and brownish-yellow in colour; m.p., 81° to 82° C.; neutralisation value, 121.8; iodine value, 4.7. The wax contained melissic acid ($C_{30}H_{50}O_2$), melissyl (myricyl) alcohol ($C_{30}H_{52}O$) and possibly saturated acids such as montanic acid ($C_{28}H_{56}O_2$) and cerotic acid ($C_{26}H_{52}O_2$). Compounds containing oxygen, which were either unsaponifiable or saponifiable with difficulty, were present in relatively large proportions. The wax could probably be used as a substitute for carnauba wax.

J. G.

Inorganic

Non-precipitation of Cobalt and Palladium by Nitro- β -Naphthol. C. Mahr and W. Prodinger. (*Z. anal. Chem.*, 1939, 117, 334.)—Methods for the gravimetric determination of cobalt and palladium by means of nitro- β -naphthol have been published by Mahr (*ANALYST*, 1934, 59, 846). It has now been found that the reagent employed in the earlier work contained large proportions of nitroso- β -naphthol, to which the precipitation of the metals was undoubtedly due; pure α -nitro- β -naphthol, prepared from β -naphthylamine, does not precipitate cobalt or palladium solutions, and therefore cannot be used for the determination of these metals.

W. R. S.

Colorimetric Determination of Iron in Aluminium and Alumina. L. Roelen. (*Z. anal. Chem.*, 1939, 117, 385–389.)—The method utilises the bluish-green colour of alkaline colloidal solutions of ferrous sulphide, which conforms to Lambert and Beer's law. It is immaterial whether the iron is present as ferric or ferrous salt; its concentration should be 0.005 to 0.3 mg. per 100 ml. A colorimetric scale is prepared by pipetting 1, 2, 3 to 10 ml. of a standard solution (0.04 mg. of Fe_2O_3 per ml.) into 100-ml. flasks, and adding 5 ml. of 5 per cent. sodium tartrate solution and 2 N sodium hydroxide solution, drop by drop, to alkaline reaction. The solution is treated with 5 ml. of 10 per cent. sodium sulphide solution; the three reagents should be freshly prepared each day. After standing for 15 minutes the solutions are diluted to 100 ml.; the colour is stable for

several hours. A Pulfrich photometer is recommended in place of the scale of standards, the colour being measured against water and light filters Hg436 or S61. *Refined aluminium* (free from lead and copper).—The sawings (1 to 2 g.) are dissolved as usual in a mixture of *aqua regia* and sulphuric acid, and the solution is heated until white fumes are evolved. Silica is filtered off, and the filtrate is diluted according to the iron-content (100 to 500 ml.). A suitable amount is treated as described above. *Alumina*.—Two g. are fused with 40 g. of potassium bisulphate in a corundum crucible (a vitreosil crucible appears more suitable—ABSTRACTOR). The cold melt is dissolved in 300 ml. of water and 50 ml. of sulphuric acid (1:1), the solution is diluted to 500 ml., and 2 to 25 ml. are submitted to the test after 30 minutes' standing; the standard should contain the same amount of bisulphate. *Hydrated alumina*.—One g. is dissolved in 10 ml. of sulphuric acid (1:1) in a 100-ml. flask, and 25 ml. are taken for the test, sufficient sodium hydroxide being added to re-dissolve the precipitated alumina. *Aluminate liquor* (2 to 5 ml.) is treated with 5 ml. of sodium tartrate solution, sulphuric acid (1:5) until the aluminium hydroxide just re-dissolves, and a slight excess of 2 *N* sodium hydroxide solution. The alkaline liquid is treated with sodium sulphide, and the tint is matched after 15 minutes.

W. R. S.

Determination of Aluminium in Presence of Iron and Phosphoric Acid. G. Balanescu and M. D. Motzoc. (*Z. anal. Chem.*, 1939, 118, 18–26.)—In the analysis of soil extracts, the basic acetate precipitate is dissolved in hydrochloric acid and the solution is made up to a definite volume. In one aliquot portion the iron is determined volumetrically. Another portion is used for the determination of phosphoric acid. To the third portion sufficient sodium phosphate is added to produce a solution in which the ratio $\text{g. Fe} : \text{g. P}_2\text{O}_5 = 2$. The liquid is then treated, drop by drop, with strong sodium hydroxide solution until the iron is completely precipitated, then with 5 ml. of *N* sodium hydroxide solution, and slowly heated to boiling. The precipitate is collected and well washed with boiling water containing 5 ml. of *N* sodium hydroxide solution per 100 ml. The filtrate is cooled and neutralised with 3 *N* hydrochloric acid to phenolphthalein; the precipitate is re-dissolved by dropwise addition of *N* sodium hydroxide solution, which restores the pink colour of the indicator. The opalescent solution (80 ml.) is cleared by boiling, cooled to 50° C., treated with a 3 per cent. alcoholic solution of 8-hydroxyquinoline, and heated to boiling. The precipitate is collected in a Jena glass filtering-crucible, G4, washed with boiling water until the washings are colourless, dissolved in 3 *N* hydrochloric acid, and titrated with 0.1 *N* bromate solution (1 ml. \equiv 0.000225 g. Al). The adjustment of the $\text{Fe}:\text{P}_2\text{O}_5$ ratio is stated to ensure the quantitative separation of iron from aluminium by a single precipitation with sodium hydroxide.

W. R. S.

Gravimetric Determination of Antimony by Means of 8-Hydroxyquinoline. T. I. Pirtea. (*Z. anal. Chem.*, 1939, 118, 26–30.)—The acid trichloride solution is treated with an excess of reagent (7 to 8 g. of the base dissolved in a minimum of acetic acid and treated with dilute ammonia till cloudy, the cloudiness being removed with a few drops of acetic acid). The liquid is heated to 70° C., and 10 per cent. ammonia is added from a pipette, a yellow precipitate beginning

to form at pH 1.5. The addition of ammonia is continued until the precipitation is quantitative and the solution becomes deep yellow. The precipitate is set aside in the cold for 1 to 2 hours, collected in a Jena glass crucible (1G3 or 1G4), washed with a solution containing 0.3 g. of the base and a few drops of acetic acid per litre, dried for an hour at 105° to 110° C., and weighed. In testing for constancy of weight, the precipitate should not be heated for more than 30 minutes at a time, as it gradually undergoes a slight loss in weight. Sb factor for $(C_6H_5ON)_3Sb$: 0.2197. Antimony may be precipitated from tartrate solution by the same method, but the precipitate is of a lighter yellow colour, and has the composition



with an antimony factor 0.2129. The antimony determination in tartar emetic can be effected in hydrochloric or tartaric acid solution; the last-mentioned basic compound is precipitated.

W. R. S.

Volumetric Determination of Chlorate and Bromate. C. Mahr and H. Ohle. (*Z. anal. Chem.*, 1939, 117, 389–391.)—The oxidation of thiourea (ANALYST, 1939, 64, 622) can be applied to this determination, the procedure having the advantage over reduction with ferrous salt that the reducing agent is stable and not affected by atmospheric oxygen. The excess of thiourea can be determined by means of permanganate in presence of iodide. *Chlorate*.—The chlorate solution is added to a mixture of 20 ml. of sulphuric acid (1:1), 5 ml. of 1 per cent. potassium iodide solution, and a measured excess of 0.1 N thiourea solution during agitation, and the liquid is heated on the water-bath to 70° C. for 10 to 15 minutes. After cooling to 35° C. it is treated with a little starch solution, diluted to 80 ml., titrated with 0.1 N permanganate to the appearance of a faint blue tint, diluted to 250 to 300 ml. with water (35° C.), and again titrated until a light blue end-point is reached. *Bromate*.—The bromate solution is added, drop by drop, during vigorous stirring, to the mixture described above. The excess of thiourea can be titrated at once with permanganate after addition of starch solution, dilution to 80 ml., and warming to 35° C.

W. R. S.

Determination of Fluorine and Silica in the Hot-Springs of Gastein. R. Bisanz and F. Kroupa. (*Chem.-Ztg.*, 1939, 63, 689–691.)—Samples from six different springs contained 1.77 to 3.40 mg. of fluorine and 35.5 to 45.4 mg. of silica per kg. of water. Fluorine was determined by titration with thorium nitrate with the use of sodium alizarinesulphonate as indicator (Armstrong, *J. Amer. Chem. Soc.*, 1933, 55, 1741) and also gravimetrically as calcium fluoride, the results of the two methods showing good agreement. Silica was determined (a) by direct separation through evaporation with hydrochloric acid; (b) by precipitation with zinc oxide and ammonium carbonate; in either method the nett weight of silica was obtained by volatilisation with hydrofluoric acid. The values were reasonably concordant, but, despite the possibility of slight loss of silica as fluoride in method (a), slightly higher results were obtained than with method (b). This point is to be further studied.

S. G. C.

Determination of Fluorine in Wood [Treated with Fluoride Preservative]. B. Ikert. (*Chem.-Ztg.*, 1939, 63, 754-755.)—It is usual to impregnate the sample of wood with calcium acetate solution before ashing prior to the determination of fluoride. It is now proposed to incorporate chromium acetate in the impregnating solution (a solution containing 6 per cent. of calcium acetate and 3 per cent. of chromium acetate is recommended.) More complete recovery of the fluorine and speedier ashing are claimed. The fluorine in the ash may be determined by distillation as silicon fluoride and subsequent titration as in Willard and Winter's method. S. G. C.

Microchemical

Spot Tests for Silver, Lead, Mercury, Uranium and Aluminium. E. A. Kocsis. (*Mikrochem.*, 1939, 27, 180-184.)—The reagents used are benzopurpurin 4B, and bromophenol blue (Merck No. 8182) in 0.2 per cent. aqueous solution and morin (Merck No. 6098) in 0.2 per cent. alcoholic solution. *Silver* nitrate and benzopurpurin 4B give a brown ring with a red spot in the middle on filter-paper; *limit of identification*, 0.03 mg. of silver. The other ions do not give this reaction. The intensity of colour is lost on drying. *Lead* nitrate and bromophenol blue form on filter-paper a violet-red circle, surrounded by and encircling a number of colours; on drying, only the circle retains its brightness; *limit of identification*, 0.025 mg. lead; silver and copper ions interfere with this test. *Mercurous* nitrate gives red-brown colours with benzopurpurin 4B and bromophenol blue, which do not fade very much on drying and may be kept for several weeks; *limits of identification*, 0.04 mg. and 0.015 mg. *Mercuric* nitrate forms blue-green and yellow ochre colours with the two above-named reagents; the colours retain their intensity for several weeks; *limits of identification*, 0.025 mg. with benzopurpurin 4B and 0.02 mg. with bromophenol blue. *Uranium* nitrate gives a light brown colour with benzopurpurin 4B and a red-brown colour with bromophenol blue. Both colours lose their intensity on drying; *limits of identification*, 0.035 mg. with benzopurpurin 4B and 0.025 mg. with bromophenol blue. *Aluminium* sulphate forms a bright red-brown colour with benzopurpurin 4B, which is unchanged by drying; *limit of identification*, 0.005 mg. of aluminium. The morin test for aluminium may be made sensitive down to 0.001% of aluminium by carrying out the test in non-fluorescent glass and observing the reaction in transmitted ultra-violet light. J. W. M.

Salts of Complex Cations applied to the Microscopic Detection of Anions. VIII. 1.2-Chloro-aquatetramminocobaltic Chloride. L. V. Yanowski and W. A. Hynes. (*Mikrochem.*, 1929, 27, 161-164.)—The reagent, $[\text{Co}(\text{NH}_3)_4(\text{H}_2\text{O})\text{Cl}]\text{Cl}_2$, was prepared by a modification of Werner's method (*Ber.*, 1907, 40, 4817), and a saturated aqueous solution of the salt was employed. Characteristic crystals are formed only with pyro- and ortho-phosphate ions, from 1 per cent. solutions of the anions. With the former pyramidal masses form immediately (*limit of identification*, 67% HPO_4^{--}); with the latter, spruce-like fronds develop in 5 to 7 minutes (*limit of identification*, 0.2 mg. $\text{P}_2\text{O}_7^{--}$). Non-characteristic crystalline precipitates are obtained with silicotungstic acid and alkali metal

salts of biniodate, dichromate and nitrite. Turbidities of varying densities are formed by phosphomolybdic and phosphotungstic acids and by dipicrylamine, as well as by the alkali metal salts of bisulphite, chromate, ferricyanide, ferrocyanide, iodate, metaphosphate, metavanadate, molybdate, orthovanadate, oxalate, paratungstate, phosphomolybdate, phosphotungstate, sulphite, tellurate, tellurite, tetraborate, tungstate, and xanthate ions. No reaction was obtained with 54 other anions that were tested. Five photomicrographs are given.

J. W. M.

Activation of Vanadium with Pyrocatechol. L. Szebelledy and M. Ajtai. (*Mikrochem.*, 1939, 26, 72-74.)—Vanadium catalyses the reaction between *p*-phenetidine and potassium bromate, and the catalysis is activated by pyrocatechol. The sample is treated with 1 ml. of a 1 per cent. solution of pyrocatechol, 1 ml. of a 0.1 per cent. solution of *p*-phenetidine in hydrochloric acid and 1 ml. of saturated potassium bromate solution. The mixture is diluted to 5 ml. and compared, after a few minutes, with a blank. In presence of 0.001 γ of vanadium the sample gives a violet colour after 5 minutes as compared with a slight pink in the blank. The limit of identification is 0.0005 γ of vanadium. The interference of silver ions may be prevented by adding 10 cg. of sodium chloride, that of lead by adding 10 cg. of sodium sulphate, and that of iron by the addition of 5 cg. of sodium difluoride.

J. W. M.

Physical Methods, Apparatus, etc.

Lead-sodium Alloy as a Drying Agent. H. Soroos. (*Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 657-658.)—Lead-sodium alloy reacts only slowly with air or water, yet dries ether as completely as sodium wire and is less hazardous to handle in presence of inflammable liquids. The alloy is very brittle and can be prepared in any desired state of subdivision. A mixture of 90 parts of lead and 10.5 parts of sodium, corresponding with the ratio NaPb that yields the lowest and safest concentration of sodium providing an active and brittle product, is heated in an iron crucible fitted with a lid through which passes an iron stirrer. After the mass is liquid the crucible is allowed to cool at an angle of 45°, and the alloy is removed and stored in an air-tight container. When required for use it is coarsely ground in a mortar, but if rapid reaction is required it may be finely ground under a layer of the liquid to be dried. Unless this precaution is taken, absorption of water from an excessively moist atmosphere may cause the finely powdered alloy to ignite. Upon reaction with water the alloy disintegrates into a fine powder, and the sodium is effectively and quantitatively used. In addition to its convenience for handling and its efficacy, it has the advantage that residues are disposed of by the addition of water without violent reaction. It is made industrially as an intermediate in the manufacture of lead-tetraethyl. A. O. J.

Reviews

THE RAMAN EFFECT AND ITS CHEMICAL APPLICATIONS. By J. H. HIBBEN, with a Theoretical Discussion by J. H. HIBBEN and E. TELLER. Pp. 544. American Chemical Society Monograph; published by Reinhold Publishing Corporation. 1939. Price 66s. net.

Since the announcement by Sir C. V. Raman in 1928 of the peculiar scattering of light rays by molecules, nearly two thousand communications on the subject of Raman spectra have been published by chemists and physicists. The universal interest aroused springs from the fact that the theoretical discussions have led to far-reaching conclusions on the structure of molecules and on molecular symmetry. The chemical application of the study may be considered quite apart from the physical processes which give rise to Raman lines. It is one of the objects of this monograph to present the facts, and deductions from them, in so far as they interest chemists. It is beyond question that the Raman effect can be correlated with certain kinds of chemical binding between atoms. Two examples, taken at random, may be given. In ethylenic compounds the $C = C$ link is associated with a frequency shift of 1600 to 1680 wave-numbers; the presence of a halogen derivative can be detected in a hydrocarbon mixture by means of the Raman shifts.

Chapters 3 to 6 deal with the theory of the Raman effect. While these are necessarily based on modern mathematical treatment, the method of presentation gives a clear picture of the processes involved, and with a little persistence it can be grasped readily. The rules which govern the appearance of Raman lines, their polarisation and the relation of these to the symmetry of the molecule are clearly set out.

For those who do not desire to study the theory but are interested in the application to inorganic and organic chemistry and to biochemistry, Parts II and III, forming the major portion of the volume, can be read independently of the theoretical aspects of Part I. For the busy chemist this is a fortunate circumstance, since he can use the book with advantage in attacking many chemical problems. For example, it is stated that the Raman spectrum shows citral to contain no α -form, whilst citronellal is a mixture of the α - and β -forms. Again, the Raman spectra of certain sugars, such as glucose, sorbose, xylose and others, do not show a Raman shift in the region 1600 to 1700 wave-numbers, and, as this is the region where the carbonyl frequency should appear, it is concluded that aldehydic or ketonic structures are absent from these substances. Hibben points out that the Raman effect offers great promise for investigating industrial problems and also in applied biochemistry. The reviewer agrees with this view, and would point out that the Raman effect and the complementary infra-red spectrum are now coming into the field of analytical chemistry. The volume is by no means difficult to read and is fully up to the high standard of this series of Chemical Monographs.

The bibliography is complete and is arranged alphabetically, but the index could be improved, in the reviewer's opinion, if a different system were adopted, namely, a general index giving page numbers. This is, however, a minor matter and in no way detracts from the excellence of the volume for study and reference.

J. J. Fox

THEORETICAL AND APPLIED ELECTROCHEMISTRY. By M. DE K. THOMPSON.
Pp. xxi + 535. 3rd Edition. New York: The Macmillan Company. 1939.
Price 22s.

The general plan of this book is similar to that of the second edition, published in 1925, but the subject-matter has been brought up to date without any expansion of the text. The first part is theoretical, and deals with electrolysis, dissociation and migration of ions, conductance, electrokinetic phenomena and polarisation. Part II includes sections on electro-analysis, plating, the extraction and refining of metals, electrolytic oxidation and reduction, the production of oxygen and hydrogen, primary and storage cells, applications of electrokinetic phenomena and corrosion. The final section is concerned with electric furnaces and their products, and includes electrothermic metallurgy. No single volume could be expected to deal extensively with each of the many applications discussed, but the author has succeeded in presenting a broad survey of electrochemical processes.

In the theoretical section facts and theories are given briefly and the reader must look up the references given in the copious footnotes in order to amplify the lecture-note type of information of the text. Indeed, these footnotes and the bibliography at the end of each chapter combine to enhance the value of this book for reference purposes.

By contrast, some sections in Part II contain a wealth of detail: dimensions of plant, operating conditions, diagrams, drawings, photographs and the like. The information was presumably made available to the author by the various companies to whom acknowledgment is made in the preface. The descriptions of some processes, for example, the manufacture of fused quartz, probably owe something to the patent literature—a source that is not always a true guide to current practice.

Descriptions of some new developments, for example the electric melting of glass, are so brief as hardly to warrant inclusion. There is a tendency to neglect sources other than American; among the tests for electroplating there is no mention of S. G. Clarke's B.N.F. Jet Test, and the information on the "salt spray" test is out of date. Much attention has been devoted to electrolytic oxidation and reduction processes in America of recent years, and a number of commercial processes are described in the book, among them the production of iodoform by oxidation and the reduction of glucose to give sorbitol and mannitol. Electrolytic analysis is allotted only six pages, and the analyst may not, therefore, expect to profit greatly from a perusal of this chapter, which deals only with general principles. The chapter on corrosion, new to this edition, begins with the statement that the American oil industry lost 175 million dollars in 1927 as a result of corrosion, but as only two pages are devoted to the subject, they might well have been omitted.

The book is well produced and misprints are few, but there is some careless usage of words. It must be one crystallite of graphite, not one molecule, that contains only 30 atoms (p. 421); electrolytic α brass has a micro-structure, not a crystal structure, different from cast brass of the same composition (p. 154).

The problems following each chapter, extended solutions to which are given at the end of the book, contribute towards an understanding of the subjects

discussed. Though primarily written for students, this book should be equally useful for reference purposes since a large part of it is devoted to practical industrial applications.

R. C. CHIRNSIDE

FLUORESCENCE ANALYSIS IN ULTRA-VIOLET LIGHT. By J. A. RADLEY and JULIUS GRANT. 3rd Edition. Pp. 424 + 28 photographs. London: Chapman & Hall. 1939. Price 22s. 6d.

Judging by the number and range of papers regularly published on fluorescence analysis there is no lack of interest in this subject. In recent years there has been no outstanding advance; but extension of application is evident in almost every branch of applied chemistry. There is thus ample justification for the appearance of this new edition which brings the subject-matter right up to date.

The general arrangement follows that of the two previous editions, accounts of which have already been recorded (*ANALYST*, 1934, 59, 209; 1936, 61, 215). Each of the first 19 chapters contains the material of the last edition (slightly revised and edited), together with additional matter published within the last few years. The final chapter is almost wholly new and comprises 17 pages dealing with recent work in the dyestuffs industries in connection with the laws of fluorescence, classification or identification of dyes, accelerated fading and the detection of faults. Nine more photographs are included.

The additions result in an increase of about one-third in the size of the book and quantitatively it may be said that 30 per cent. more pages have been provided for a 7 per cent. increase in price. Yet qualitatively the new material is more valuable than the old, for exaggerated claims, referred to in the review of the last edition as occurring in much early work, have been succeeded by more cautious estimates.

The authors have now on three occasions provided an admirable, up-to-date collection of the published work on fluorescence analysis. The latest volume approaches the limiting size for convenience, and for a subsequent edition it may be necessary to delete some of the information and references now included to make room for newer work. The task of separating the chaff from the grain will be difficult; but it is hoped that the authors will not shirk this winnowing, as it would improve the harvest for the increasing number of people who utilise the seed that has been garnered for them.

J. R. NICHOLLS

TECHNICAL METHODS OF ORE ANALYSIS FOR CHEMISTS AND COLLEGES. By A. J. WEINIG and W. P. SCHODER. Based upon the text by A. H. Low. Eleventh Edition. Pp. x + 325. New York: John Wiley & Sons, Inc.; London: Chapman & Hall, Ltd. 1939. Price 22s. 6d.

All analysts engaged in metallurgical practice will welcome the new edition of the late Dr. Low's well-known manual, revised by A. J. Weinig, Director, and W. P. Schoder, Chemist, of the Experimental Ore Dressing and Metallurgical Plant of the Colorado School of Mines.

The greater part of the text is a revised and up-to-date reproduction of Dr. Low's excellent work, and since this is (or should be) on the bookshelf of every metallurgical laboratory, further commendation appears superfluous. The reviewer's remarks are therefore confined to the new features of the present edition.

The introductory chapters of the former editions on apparatus, electrolysis, and logarithms have been omitted and replaced by a 24-page chapter entitled "Semi-micro Methods." This consists of a collection of 42 qualitative spot tests for the most important elements, preceded by an illustrated description of the apparatus required, and supplemented by two handsome coloured plates showing the result of the colour test in each case. These tests are intended to be carried out on a drop withdrawn from the assay solution.

One new chapter—on beryllium—has been added to those devoted to the quantitative determination of the various elements. The reviewer is gratified to find that the method described by the authors for the determination of beryllium is based on the process he advocated as the simplest for the separation of beryllium from aluminium, namely, fusion of the ammonia precipitate with sodium carbonate (ANALYST, 1936, 61, 235).

Each of the chapters on individual elements now comprises a table giving the most important minerals of the element under discussion, with useful data on properties and associated elements and reagents available for their decomposition. The table on cadmium minerals cites only the rare mineral greenockite; in the reviewer's opinion, it would be preferable to include zinc blende which, though not a cadmium mineral, is the only cadmium ore of commercial importance. Again, the table on silicate minerals does not mention quartz, by far the most abundant source of silica. Zinc blende might have been added to the sulphur minerals, since it is extensively used for the manufacture of sulphuric acid.

It may be suggested that an account of the colorimetric determination of small amounts of bismuth would form a most useful addition to the bismuth chapter.

Among the few misprints that have escaped proof-reading may be noted the atomic weight of hydrogen, in the table inside the back-cover (1.081 for 1.0081); and the gravimetric factors for zirconium pyrophosphate (p. 308). The formula of this compound should read ZrP_2O_7 (not $\text{Zr}_2\text{P}_2\text{O}_7$), and the factors for Zr and ZrO_2 be corrected accordingly.

W. R. SCHOELLER

A TEXT-BOOK OF QUANTITATIVE INORGANIC ANALYSIS. Theory and Practice. By ARTHUR I. VOGEL, D.Sc., D.I.C., F.I.C. Pp. xix + 856. With 4 plates and 130 diagrams. London: Longmans, Green & Co. 1939. Price 18s.

In this book the balance between classical methods, of importance for their instructional value, and those required in commercial work, is struck and maintained in a way that should appeal both to teachers and to those engaged in the practice of analytical chemistry.

In the theoretical section (193 pp.) general theory receives full mathematical treatment and is illustrated by fully-worked examples of the several laws. This is followed by the theory of volumetric and gravimetric analysis, the subject being taken from first principles to its most recent stage of development. Technique (84 pp.) is treated first in general, and then with detailed description of the apparatus, calibrations and manipulation required for volumetric and gravimetric analysis. The chapter on volumetric analysis (195 pp.) contains, in addition to the usual elementary matter, descriptions and exercises on adsorption indicators and methods utilising mercurous perchlorate, ceric sulphate, manganic sulphate, titanous sulphate, liquid amalgams and chloramine-T.

The section on gravimetric analysis (197 pp.) covers simple determinations, systematic analysis (in which 13 of the commoner "rare" elements have not been overlooked), electrolytic determinations, simple separations and analysis of complex materials such as alloys and minerals. The critical discrimination and modern outlook of much of this and the preceding section bear more resemblance in style to that of a work of reference than a text-book; this adds interest for the chemist engaged in general analysis. In selecting methods of determination full use has been made of recently published original papers, including some from the pages of *THE ANALYST*. For all elements a wide choice of procedure is offered.

Colorimetric Analysis receives 62 pages, of which 29 are devoted to theory and instruments; procedures are described for 19 characteristic ions, and a table of reference is provided for 26 other elements and radicals. Gas Analysis (46 pp.) describes and illustrates the use of the Hempel, Bunte, Orsat, Ambler, and Bone-Wheeler apparatus and the Lunge nitrometer. An appendix (65 pp.) contains, in addition to all the usual matter, a table of buffer solutions for standardisation of *pH* measurements, a comprehensive bibliography of analytical chemistry and a table of *five-figure* logarithms. The index (14 pp.) is rendered easy of reference by heavy type for main headings and a system of abbreviations to indicate the class to which a determination belongs.

This work, which is remarkably free from errors, is likely to set a new standard for text-books of quantitative inorganic analysis.

F. L. OKELL

Publications Received

PHYSICO-CHEMICAL METHODS. Vol. I. MEASUREMENTS AND MANIPULATIONS. Vol. II. PHYSICAL PROPERTIES. 3rd Ed. By J. REILLY and W. NORMAN. London: Methuen & Co. 1940. Price £4 4s. net.

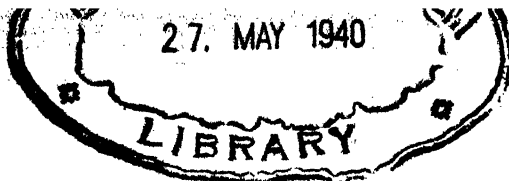
AN INTRODUCTION TO BIOCHEMISTRY. 2nd Ed. By W. R. FEARON. Pp. xii + 475. London: William Heinemann (Medical Books), Ltd. 1940. Price 17s. 6d. net.

PRACTICAL PHARMACEUTICAL CHEMISTRY. 4th Ed. By F. N. APPLEYARD and C. G. LYONS. Pp. viii + 174. London: Sir Isaac Pitman & Sons, Ltd. 1939. Price 6s. 6d. net.

METALLURGICAL ANALYSIS AND ASSAYING. By J. S. REMINGTON and F. L. JAMESON. Pp. vii + 101. London: The Technical Press, Ltd. 1939. Price 5s. net.

SULPHATED OILS AND ALLIED PRODUCTS. Pp. 167. London: A. Harvey. 1939. Price 12s. 6d. net.

LES PARFUMS NATURELS. By Y. R. NAVES and G. M. MAZUYER. Pp. xvi + 398. Paris: Gautiers Villars. 1939. Price 120 fr.



THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

THE Annual General Meeting of the Society was held in the Chemical Society's Rooms, Burlington House, W.1, on Wednesday, March 6th, at 2.50 p.m., the President, Professor W. H. Roberts, being in the chair. The Honorary Treasurer presented the accounts for the year, and the Honorary Secretary presented the Annual Report of the Council. The President delivered his Presidential Address.

The following were elected as Officers and Council for the year 1940:

President.—E. B. Hughes, D.Sc., F.I.C.

Past Presidents serving on the Council.—F. W. F. Arnaud, Bernard Dyer, John Evans, Edward Hinks, G. Roche Lynch, W. H. Roberts, G. Rudd Thompson.

Vice-Presidents.—E. B. Anderson, S. E. Melling, F. G. H. Tate, J. R. Stubbs (*Chairman, North of England Section ; ex officio*), T. Cockburn (*Chairman, Scottish Section ; ex officio*).

Honorary Treasurer.—G. Taylor.

Honorary Secretary.—Lewis Eynon.

Other Members of Council.—C. A. Adams, F. C. Bullock, H. E. Cox, D. C. Garratt, L. H. Lampitt, G. W. Monier-Williams, C. J. Regan, T. Rendle, A. Scholes, W. M. Seaber, W. H. Simmons, W. W. Taylor, J. B. McKean (*Honorary Secretary, Scottish Section ; ex officio*), A. Lees (*Honorary Secretary, North of England Section ; ex officio*).

The following candidates have been elected members of the Society:

Aage Jorgen Christian Andersen, B.A., M.Sc. (Copenhagen).

Isaac Berg, B.Sc. (Lond.), A.I.C.

William Edwin Joseph Richard Calcutt, B.A., D.I.C., A.I.C.

Richard Arthur Dalley, A.I.C. (*Through North of England Section.*)

William Thomas Elwell, A.I.C. (*Through North of England Section.*)

Joseph Markland, B.Sc. (Lond.), A.I.C. (*Through North of England Section.*)

Cecil Hancorn Robins, B.Sc. (Lond.), A.I.C.

Robert Orr Scott, B.Sc., A.R.T.C., A.I.C. (*Through Scottish Section.*)

Joseph Henry Singer, A.I.C.

George Edgar Turfitt, B.Sc., Ph.D. (Lond.), A.I.C. (*Through North of England Section.*)

SCOTTISH SECTION

THE Fifth Annual General Meeting of the Section was held in the Bath Hotel, 154, Bath Street, Glasgow, on February 22nd, 1940.

The Chairman, Mr. T. Cockburn, gave an appreciation of the late Dr. T. W. Drinkwater, who had been Vice-Chairman of the Section during 1935 and 1936.

The Secretary read the Report and Financial Statement for 1939, which were adopted.

The following Office-bearers were elected:

Chairman.—T. Cockburn; *Vice-Chairman.*—J. W. Hawley; *Committee.*—J. Brown, A. Dargie, A. R. Jamieson, R. G. Thin, R. T. Thomson and J. F. Tocher; *Honorary Secretary and Treasurer.*—J. B. McKean; *Honorary Auditors.*—M. Herd and R. S. Watson.

The following papers were read and discussed:—"The Pollution of Water by Fish," by R. T. Thomson, F.I.C., and "Notes on some War Gases: The Detection of Mustard Gas in Food," by J. B. McKean, F.I.C., and J. A. MacNair, F.I.C.

Death

WITH great regret we record the death, on January 9th, of George Arthur Pingstone, Public Analyst for the Municipality of Bulawayo. He had been a member of the Society for 36 years.

Annual Report of Council

MARCH, 1940

THE Roll of the Society numbers 886, an increase of 29 over the membership of last year.

The Council regrets to have to record the death of the following members:

W. T. Burgess
R. A. Cripps
T. W. Drinkwater
F. G. Edmed
D. L. Howard
H. F. E. Hulton
H. T. Lea
Sir G. T. Morgan
G. A. Pingstone
Sir W. J. Pope
F. T. Shutt
K. E. N. Williams

and of G. Stubbs, a former member.

Burgess, who died at the age of 78, had been a member of the Society for 40 years and served on the Council as an Ordinary Member and as a Vice-President. During almost the whole of his career he was engaged in work connected with water supply, first as an assistant to Sir Edward Frankland, then as Water Analyst to the Local Government Board, and finally as a private practitioner. (Obituary, ANALYST, 1940, 65, 1.)

Cripps, who died at the age of 76, had been a member of the Society for 43 years, and served as a member of Council in 1910-11. He was for many years Public Analyst for Bournemouth. His work was chiefly in connection with pharmaceutical chemistry, and he published numerous papers on pharmaceutical subjects. (Obituary, ANALYST, 1939, 64, 394.)

Drinkwater, who died in his 90th year, had been a member of the Society for 16 years. He was for over 60 years a lecturer in the School of Medicine of the Royal College of Physicians and Surgeons, Edinburgh, and held a number of appointments as Public Analyst.

Edmed, who died at the age of 62, had been a member of the Society since 1930, and served as a Member of the Council in 1934-35. He was for many years Admiralty Chemist at H.M. Dockyard, Portsmouth.

Howard, who died at the age of 73, had been a member of the Society since 1900, and served on the Council in 1906-07 and 1918-19. He was a Director of Messrs. Howards & Sons, and was for many years a Justice of the Peace for Essex.

Hulton, who died at the age of 64, was elected a member of the Society in 1913, and served as a Member of Council in 1923-24. Practically the whole of his professional life was devoted to the brewing industry, and in collaboration with J. L. Baker he published a number of papers on brewing subjects. (Obituary, ANALYST, 1939, 64, 639.)

Lea, who died in his 50th year, had been a member of the Society since 1919, and served on the Council 1926-27. During the war of 1914-18 he served in the Special Brigade, Royal Engineers. He subsequently became a Consulting Chemist and held appointments as Public Analyst and Official Agricultural Analyst. (Obituary, ANALYST, 1939, 64, 778.)

Sir G. T. Morgan, who died in his 70th year, was an Honorary Member of the Society. After a long and distinguished academic career, during which he made many important contributions to pure and applied chemistry, he became, in 1925, Director of the Chemical Research Laboratory at Teddington, and held that position until 1937, after which he became the Research Director of the Institute of Brewing. He was awarded the honour of Knighthood in 1936.

Sir W. J. Pope, who died in his 70th year, had been a member of the Society since 1915. He was for over 30 years Professor of Chemistry at Cambridge. His discovery of the optical activity of compounds of nitrogen, sulphur, selenium and tin gave him an international reputation as a research worker in the field of pure chemistry. During the war of 1914-18 he developed a process for the synthesis of mustard gas far in advance of that used in Germany. (Obituary, ANALYST, May, 1940.)

Shutt, who died in his 81st year, became a member of the Society in 1916. He had a long and distinguished career in the field of agricultural chemistry in Canada, and held the post of Dominion Chemist in the Department of Agriculture, Ottawa. He was awarded the honour of C.B.E. in 1935.

Stubbs, who died at the age of 75, was elected a member of the Society in 1926, and served on the Council as an Ordinary Member and as a Vice-President. He was for many years in the Government Laboratory, finally as Deputy Government Chemist.

BIENNIAL LECTURE.—Following the Annual General Meeting on March 3rd, 1939, the Biennial Lecture was given by Sir Henry Dale, C.B.E., M.D., F.R.S., the subject being "Biological Standardisation" (ANALYST, 1939, 64, 554).

ORDINARY MEETINGS.—Owing to the outbreak of war it was found necessary to cancel the October and November meetings. During the year, five meetings were held, and the following papers were communicated:

- "Food Inspection and Analysis in Holland." By J. Straub, Chem. Ing., Director of the Food Inspection Laboratory for Amsterdam.
- "The Determination of Traces of Zinc in Biological Material and Natural Waters." By Noel L. Allport, F.I.C., and C. D. B. Moon, A.I.C.
- "The Evaluation of Hydrogen Peroxide." By Mrs. S. M. L. Tritton, M.P.S., F.I.C.
- "Extract of Malt with Cod-liver Oil: Determination of Oil and Vitamin A." By D. C. Garratt, B.Sc., Ph.D., F.I.C.
- "The Presence of Leuco-anthocyanins in Criollo Cacao." By the late A. W. Knapp, M.Sc., M.I.Chem.E., F.I.C., and J. F. Hearne, A.I.C.
- "Notes on the Examination of Textiles in Cases of Suspected Dermatitis." By H. E. Cox, Ph.D., D.Sc., F.I.C.
- "The Examination of Lard." By R. W. Sutton, B.Sc., F.I.C., A. Barraclough, B.Sc., F.I.C., R. Mallinder, B.Sc., F.I.C., and O. Hitchen, B.Sc., F.I.C.
- "The Estimation and Examination of 2-Methyl-1:4-naphthoquinone." By J. L. Pinder, B.Sc., F.I.C., and J. H. Singer, A.I.C. With Introductory Remarks on the Relationship of 2-Methyl-1:4-naphthoquinone to Vitamin K, by A. L. Bacharach, M.A., F.I.C.

The February meeting was a joint meeting with the Food Group of the Society of Chemical Industry, at which the following papers on Carotene and Allied Pigments were presented:

- "The Constitution and Physiological Significance of Carotene and Allied Pigments." By R. A. Morton, D.Sc., F.I.C.
- "The Commercial Determination of Carotene and Allied Pigments, with especial reference to Dried Grass and other Leafy Materials." By W. M. Seaber, B.Sc., F.I.C.

THE ANALYST.—Our journal continues to reflect the satisfactory position of the Society. It was feared at first that the outbreak of war would have a serious effect on the number of papers submitted for publication. Happily this expectation has not been realised, and the Editor is still receiving numerous contributions. By a coincidence the number of pages (924) in *THE ANALYST* was exactly the same as in the preceding year. The total number of papers published was 48, and these may be roughly classified into 15 on food and drugs, 15 inorganic, 6 organic, 5 biological, 2 on water, 3 on gas analysis, 1 agricultural, and 1 forensic. It will be noted with satisfaction that our journal is now recognised as a suitable medium for the publication of papers on inorganic analysis and metallurgy. In addition to the papers there were also 38 notes on subjects of analytical interest, and the usual abstracts from the reports of Government Analysts and Public Analysts. Publishers, as well as readers of the journal, still value the critical reviews published in *THE ANALYST*, for no fewer than 72 books were reviewed in the course of the year.

HON. TREASURER'S REPORT.—The Hon. Treasurer reports that the financial position of the Society continues to be satisfactory; the year's income has more than balanced the year's expenditure. It is hoped that no need will arise for increasing the subscription beyond one guinea, the traditional subscription to the Society.

EMERGENCY COMMITTEE.—At the time of the international crisis in September, 1938, an Emergency Committee was formed to act for and on behalf of the Council in a state of national emergency. On the outbreak of war in September, 1939, the Committee met and decided that, in view of the impossibility of holding

meetings of the Society in the near future and the uncertainty as to the holding of meetings subsequently, the procedure for the election of new members should be temporarily as follows:

- (a) All applications for membership shall first be considered by the Committee.
- (b) Those approved shall be circulated for comment to all members of the Council.
- (c) The Committee shall reconsider them with the comments received, if any, and decide finally which applicants shall be admitted to membership.

FOOD AND DRUGS ACT, 1938.—Draft Regulations for bread and flour and applications for regulations for cheese, cream, ice-cream, honey, vinegar, coffee and coffee mixtures, have been subjects of correspondence between the Ministry of Health and the Society. Shortly after the outbreak of war, however, the Ministry informed the Society that the consideration of these draft regulations and applications must be deferred for the time being.

CENTRAL REGISTER FOR NATIONAL SERVICE.—The Society has co-operated with other scientific and professional bodies in supplying the Ministry of Labour with the names and qualifications of persons willing to be enrolled in the Central Register for National Service.

SOCIETY'S ARCHIVES.—Minutes of meetings of Council and Committees and other important records and a set of early numbers of *THE ANALYST* have been sent to Messrs. W. Heffer & Sons, at Cambridge, for safe storage against war risks, and the Council desires to express thanks to Messrs. Heffer for this accommodation.

METROPOLITAN WATER BOARD BILL.—The Society presented a Petition against the Metropolitan Water Board Bill, and in particular against Clause 51 of the Bill whereby the Metropolitan Water Board would be empowered to establish a general practice in water and sewage and effluent analysis and examination extending over the whole of the country and to receive remuneration for such practice.

The Petition was successful, Clause 51 being deleted from the Bill. The Council desires to record its thanks to the Institute of Chemistry for supporting the Petition and for defraying part of the cost of the Petition.

ANALYTICAL METHODS COMMITTEE.—Two reports from the Committee have been published during the past year, *viz.*:

"Determination of Copper in Food Colouring Materials." *ANALYST*, 1939, **64**, 339.

"Assay of Lobelia." *ANALYST*, 1939, **64**, 581.

A new Sub-Committee has been appointed, under the chairmanship of Dr. E. B. Hughes, to consider methods for determination of dry solid matter and of copper in tomato purée and other tomato products.

The work of the Sub-Committees has been seriously interfered with by war conditions, and in some cases has had to be suspended for the time being.

The Council appointed Mr. J. R. Nicholls to the Committee in place of the late Mr. E. R. Bolton, and has also appointed Mr. N. Evers an additional member.

ANALYTICAL INVESTIGATION SCHEME.—One research ("Manganese and Caffeine Contents of some Teas and Coffees") by Dr. R. K. Coleman and F. G. Gilbert has been completed and published in *THE ANALYST* (1939, **64**, 706), and three problems are still under investigation. Discussions have been in progress with a view to extending the usefulness of the Scheme by increased co-operation with the chemical departments of different universities and colleges; it is hoped that as conditions become more settled these discussions will be resumed.

NORTH OF ENGLAND SECTION.—Owing to the war the usual number of five meetings has been reduced to three. Six papers have been read:

“Silicosis and the Analyst.” By Dr. F. S. Fowweather, M.Sc., Ch.B., F.I.C., D.P.H.

“Some Notes on the Examination of Paraldehyde.” By A. R. Tankard, F.I.C., D. J. T. Bagnall, A.C.G.F.C., F.I.C., and A. Smith, B.Sc., F.I.C.

“An Improved Table for use with the British Standard Specification Hydrometers for Milks, B.S.S., No. 734—1937.” By J. G. Lunt, B.Sc., F.I.C.

“Lead in Food.” By G. W. Monier-Williams, O.B.E., M.C., M.A., Ph.D., F.I.C.

“Some Aspects of the Purification of Polluted Waters for Industrial Use.” By J. G. Sherratt, B.Sc., F.I.C.

“The Estimation of Lead in Drinking Waters.” By C. H. Manley, M.A., F.I.C.

The Annual Address by the Chairman.

There has been a good attendance at the meetings, and the President has attended.

The tenth Summer Meeting, held at Scarborough, was as successful as former ones. Dr. G. W. Monier-Williams gave the Paper, which was followed by a very interesting discussion. The number attending the meeting was above the average for previous years.

Eight candidates have applied for membership of the Parent Society through the Section.

The Section now numbers 130 members, an increase of seven on the previous year.

The Honorary Secretary wishes to acknowledge with thanks the loyal support and help of the Chairman, Officers and members of the Committee in carrying out the work of the Section.

SCOTTISH SECTION.—Two meetings were held in the year, the usual November meeting being cancelled owing to war-time difficulties of travelling and lighting.

The following papers were read and discussed.

“Sampling.” By W. M. Cameron.

“Seasoning Materials.” By R. H. McKinlay, F.I.C.

“Counterfeit Coins.” By A. Scott-Dodd, B.Sc., Ph.D., F.I.C.

“The Assay of Mercury.” By James Sandilands, F.I.C.

“Description of a Closed Respiration Apparatus for Plant Tissue.” By A. M. Smith, Ph.D., D.Sc., A.I.C.

Two new members joined the Parent Society through the Section and one member resigned.

With the approval of the Council, a Memorandum was sent to the Local Government and Public Health Consolidation (Scotland) Committee requesting the introduction into Scotland of a Food and Drugs Act on similar lines to the Food and Drugs Act, 1938. A formal acknowledgment of the Memorandum was received.

CONGRESSES.

The Society was represented by the President and Mr. A. L. Bacharach at a meeting on the subject of “Milk in its Nutritional Aspects” convened by the British Association.

Royal Sanitary Institute Congress, Scarborough, 1939.—Mr. A. R. Tankard represented the Society at the Congress of the Royal Sanitary Institute at Scarborough.

Congress of Industrial Chemistry, Warsaw, 1939.—Dr. L. H. Lampitt was appointed to represent the Society at the 19th Congress of Industrial Chemistry, Warsaw. Owing to the outbreak of war, however, the Congress was cancelled.

British Standards Institution.—The President was appointed to represent the Society on the Chemical Divisional Council of the British Standards Institution in place of the late Mr. E. R. Bolton.

The Council desires to record its thanks to the members of the Society who have served on the Committees and Sub-Committees and to those who have represented the Society on other bodies, and to various organisations and members of the Society who have afforded accommodation and hospitality to the Committees.

W. H. ROBERTS, *President*
LEWIS EYNON, *Hon. Secretary*

The Determination of small amounts of Formaldehyde in Air

By R. W. KERSEY, B.Sc., J. R. MADDOCKS, A.I.C., AND
T. E. JOHNSON, A.I.C.

THE health hazards arising from formaldehyde are known to be serious. Information published by the Association of British Chemical Manufacturers¹ indicates that the limit of toleration of formaldehyde in air is of the order of 0.025 mg. per litre. Accordingly the authors were led to devise a method for the determination of formaldehyde when present in air at such a low concentration. The quantities involved were so small as to preclude the use of methods normally available for concentrated solutions. The following methods were examined:—

A. *Absorption in iodine solution (Romijn's Method²).*—Solutions were prepared by mixing equivalent amounts of standard (N/10) iodine solution and sodium hydroxide solution and diluting to a known volume with water. Known volumes of air under test were aspirated through portions of the solution and the change in iodine-content was determined. Very low figures were obtained, corresponding to 0.0003 mg. of formaldehyde per litre of air, even in an atmosphere artificially charged by contact with 40 per cent. formaldehyde solution.

B. *Absorption in water followed by colorimetric determination.*—The usual type of cylindrical bubbler, with the inlet tube drawn out to a fine orifice and the cylinder packed with glass beads to give small bubbles and provide an increased surface for absorption, gave unsatisfactory results. The concentration of formaldehyde in the water failed to "build up" with increasing volumes of air.

A Jena glass distribution tube, with fused-in fritted glass disc (Type 33 c. G.1; diameter of pores 100–120 microns), which divided the air into a fine gas spray, was next tried. An atmosphere was artificially produced, having such a high concentration of formaldehyde that it caused acute discomfort; its formaldehyde-content must therefore have approached 0.025 mg. per litre (*cf.* Ref. 1). When this atmosphere was passed through water *via* the glass distributor tube, figures obtained were of the order of only 0.003 to 0.004 mg. per litre. With two absorbers in series, the first containing 600 ml. of water and the second 150 ml., the

concentration in the first absorber rose to 10 parts per 1,000,000 without any being detectable in the second vessel, the test used (Schryver's method, see later) being sensitive to 0.5 part per 1,000,000. Mechanical loss by non-absorption was therefore unlikely, and a possible explanation of the low results was the formation of oxidation products due to passing a continuous air stream through the solution containing absorbed formaldehyde.

C. *Application of Schryver's Method.*³—This method depends on the formation of an intense magenta colour, when dilute solutions of formaldehyde phenylhydrazones are treated with potassium ferricyanide in presence of an excess of hydrochloric acid, the depth of colour, between certain limits, being proportional to the concentration of formaldehyde present. Examination of the method, using solutions of formaldehyde of known concentration, prepared by triple dilution of the 40 per cent. aqueous solution, indicated that the working range of the test was approximately 1 to 5 parts of formaldehyde per million of solution, though 0.5 p.p.m. was detectable. Above 5 p.p.m. the colour was too intense for accurate evaluation.

The possibility of absorbing the formaldehyde from the atmosphere in a dilute aqueous solution of phenylhydrazine hydrochloride seemed to offer a method free from the disadvantages associated with absorption in water alone.

TABLE I

| Ref. No. | Volume of air, litres | Duration of test, hours | Formaldehyde in test solution p.p.m. | Concentration of formaldehyde in atmosphere, mg. per litre | Remarks |
|----------|-----------------------|-------------------------|--------------------------------------|--|---|
| 1 | 22 | 3 | 6 | 0.014 | Atmosphere too strong for continuous breathing. |
| 2 | 10 | 1 | 3 | 0.015 | Ditto. |
| 3 | 14½ | 2 | 5 | 0.017 | Formaldehyde distributed on shed floor. |
| 4 | 29 | 4 | 1 | 0.0016 | Slight smell of formaldehyde. |
| 5 | 34 | 3½ | 2 to 3 | 0.0037 | Stronger than 4. |
| 6 | 41 | 3½ | 1 | 0.0012 | Test done after running fan for ½ hr. after test 3. |
| 7 | 35 | 2½ | 0.5 to 1 | 0.001 | Test after running fan for 2 hrs. after test 5. |

A solution of phenylhydrazine hydrochloride was prepared by adding 2 ml. of conc. hydrochloric acid to a mixture of 1 g. of phenylhydrazine with water, making up to 100 ml. with water and filtering from traces of suspended matter.

Ten ml. of this solution were diluted with 50 ml. of water, and air free from formaldehyde was drawn through a Jena glass distribution tube (0.75 inch diameter) set in an absorption bottle of slightly larger diameter; the rate of flow was approximately 10 litres per hour. Slight coloration of the solution began to take place after the passage of 40 litres air. In subsequent tests, therefore, the volume of air was kept below this figure whenever feasible.

When air containing formaldehyde was passed through this solution, according to the method described later, figures for formaldehyde-content were in

direct proportion to the volume of air used, *i.e.* the formaldehyde "built up" in solution, in contradistinction to the experiments in which water only was used.

When two absorption bottles in series were used, each having a Jena glass distribution tube, no formaldehyde was detected in the solution from the second absorption bottle, showing that complete absorption was taking place in the first bottle.

The results obtained are summarised in Table I. Although the accuracy of the method has not been verified formally by analysis of mixtures of air and formaldehyde of known composition, there is every reason to suppose that the results obtained are correct within the usual limits of error of a colorimetric method.

METHOD.

SPECIAL REAGENTS REQUIRED.—(1) *Phenylhydrazine hydrochloride solution*, 1 per cent. Suspend 1 g. of phenylhydrazine in about 5 ml. water, add 2 ml. of concentrated hydrochloric acid (sp.gr. 1.16) and dilute to about 80 ml. Filter off any insoluble matter and dilute the filtrate to 100 ml. with water.

(2) *Standard formaldehyde solution.*—*Solution A.*—Dilute 25 ml. of 40 per cent. w/v formaldehyde to a litre with water (1 ml. \equiv 0.01 g. of formaldehyde). *Solution B.*—Dilute 10 ml. of Solution A to a litre with water (1 ml. \equiv 0.0001 g. of formaldehyde). *Solution C.*—Dilute 10 ml. of Solution B to a litre with water (1000 ml. \equiv 1 mg. of formaldehyde).

If desired, a stronger Solution C may be used by diluting a larger volume of Solution B to a litre with water, *e.g.* 50 ml. of Solution B diluted to a litre will give a solution containing 5 mg. of formaldehyde per litre. Solutions A, B, and C must be made freshly when required.

PROCEDURE.—Into a cylindrical wash-bottle fitted with a Jena glass distribution tube (diameter, 0.75 in.) with fused-in fritted glass filter disc (Type 33 c. G.1—pore diameter 100 to 120 microns) place 10 ml. of the phenylhydrazine hydrochloride solution and dilute to 50 ml. with water. Aspirate through this solution, at a rate of about 10 litres per hour, such a volume of air to be tested that the final solution will contain between 1 and 5 mg. of formaldehyde per litre. (The volume of air used should not exceed 40 litres.)

Dilute the solution to 50 ml. (Test Solution). Transfer 10 ml. of the Test Solution to a 20-ml. Nessler cylinder, add 1.0 ml. of potassium ferricyanide solution (5 per cent., freshly prepared) and 4.0 ml. of conc. hydrochloric acid (sp.gr. 1.16), dilute to 20.0 ml. with water, mix well and allow to stand for ten minutes.

Prepare simultaneously a series of standard comparison solutions in similar Nessler cylinders, using 2 ml. of the phenylhydrazine hydrochloride solution (reagent 1), 7 ml. of water, measured amounts of Solution C (say, 1 ml., 2 ml., 5 ml.), 1.0 ml. of potassium ferricyanide solution (5 per cent.) and 4.0 ml. of conc. hydrochloric acid (sp.gr. 1.16); dilute to 20.0 ml. with water, mix each well and allow to stand for ten minutes. Compare the colour of the Test Solution against those of the standard comparison solutions.

We wish to thank Imperial Chemical Industries, Ltd., for permission to publish this paper.

REFERENCES

1. Association of British Chemical Manufacturers Quarterly Summary, October-December, 1934. No. 2 (Vol. 5), p. 58. List of harmful concentrations of various gases.
2. *Z. anal. Chem.*, 1897, 36, 9. Abst., *ANALYST*, 1897, 22, 221.
3. S. B. Schryver, "On the Application of Formaldehyde to Meat." Food Report No. 9, 1909. Reports of the Local Government Board on Public Health and Medical Subjects. New Series, No. 12.
See also E. H. Callow, *ANALYST*, 1927, 52, 391. "The Presence of Formaldehyde in Wood Smoke and in Smoked Foodstuffs."

December 17th, 1939

The Determination of Members of the Sulphanilamide Group of Drugs

By G. V. JAMES, M.Sc., F.I.C.*

BIOCHEMICAL control during and after use of drugs of the sulphonamide group is essential to secure a concentration in the blood that is bacteriologically effective and yet sufficiently low for no toxic effects to be produced; such effects, however, are difficult to predict purely from the known drug concentration in the blood, since individuals vary in their reaction to the drug.

Probably the two most widely used members of the class are sulphanilamide itself and sulphanilamido-pyridine, but there are many other substances which owe their activity to the liberation of sulphanilamide in the body; some of these are included here, since they are still sold and may be used should a temporary shortage of the more active members of the class arise.

The sulphanilamide class may be divided broadly into those in which a substituent is introduced into the amino group, such as prontosil, supron, etc., and those in which the substituent is introduced into the sulphonamide group. Moreover, the body helps to rid itself of the drugs by acetylation of the amino group and by oxidation; oxidation applies more particularly to sulphanilamide than to sulphanilamido-pyridine, which seems only to be acetylated. The determination of the acetyl derivatives and oxidation products, however, is important, as it enables an estimate to be formed of the quantity of drug absorbed, and hence potentially active.

Table I shows the fate of many of these drugs, so far as is at present known, in the human subject, and of some of them in mice.

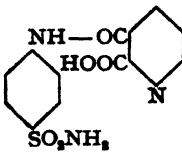

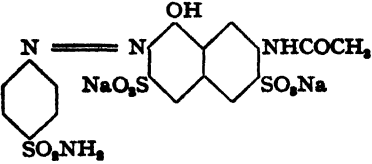
The results in Table I are averages, the number of experiments being shown in brackets; the individual figures often differ widely, as is to be expected in biological work. The experiments on mice were made with at least 8 animals in a metabolism cage, from which the excreta could be collected.

SULPHANILAMIDE (Streptocide, Ambeside, Prontylin, Prontosil Album) is a white crystalline powder (m.p. 165-166.5° C.), slightly soluble in cold water (to

* Working under a full-time grant from the Medical Research Council.

form a neutral solution), soluble in hot water, hot alcohol and cold acetone, slightly soluble in ether, benzene or chloroform, and soluble in acid or alkaline solution. In acetic acid solution it condenses with an alcoholic solution of xanthydrol to

TABLE I
THE FATE OF VARIOUS DRUGS OF THE SULPHANILAMIDE GROUP

| Drug | Formula | Animal | Urine | | | |
|---------------------------------------|---|----------------------|-----------------|------------------|------------------|-----------------|
| | | | Total Per Cent. | Oxidiz Per Cent. | Faeces Per Cent. | Total Per Cent. |
| Sulphanilamide | $\text{NH}_2\text{C}_6\text{H}_4\text{SO}_2\text{NH}_2$ | Mouse (4) Man (5) | 44.2 48.8 | 2.9 17.2 | 0.0 0.0 | 47.1 66.0 |
| Sulphanilamido-pyridine | $\text{NH}_2\text{C}_6\text{H}_4\text{SO}_2\text{NHC}_5\text{H}_4\text{N}$ | Mouse (2) Man (3) | 80.9 78.6 | 0.0 0.0 | 8.4 15.6 | 89.3 94.2 |
| Supron |  | Man (2) | 50.7 | — | 0.0 | 50.7 |
| Uleron | $\text{NH}_2\text{C}_6\text{H}_4\text{SO}_2\text{NHC}_6\text{H}_4\text{SO}_2\text{N}(\text{CH}_3)_2$ | Mouse (1) | 75.0 | 0.0 | 16.7 | 91.7 |
| Ilvin | $\text{NH}_2\text{C}_6\text{H}_4\text{SO}_2\text{NHC}_6\text{H}_4\text{COCH}_3$ | Mouse (2) | 12.3 | — | 55.0 | 67.3 |
| Benzyl sulphanilamide or proseptasine | $\text{C}_6\text{H}_5\text{CH}_2\text{NHC}_6\text{H}_4\text{SO}_2\text{NH}_2$ | Mouse (3) Man (3) | 1.9 17.8 | — — | 87.0 — | 88.9 17.8 |
| Soluseptasine | $\text{C}_6\text{H}_5\text{CH}(\text{SO}_2\text{Na})\text{CH}_2\text{CH}(\text{SO}_2\text{Na})\text{NHC}_6\text{H}_4\text{SO}_2\text{NH}_2$ | Mouse (1) | 98.5 | 0.0 | 0.0 | 98.5 |
| Prontosil rubrum* |  | Mouse (1) | 9.3 | — | — | 9.3 |
| Prontosil soluble* |  | Mouse (1) | 55.0 | — | — | 55.0 |

* The results for the two prontosils are taken from Fuller's paper.¹

form a crystalline compound (m.p. 209° C.); on this reaction has been based a gravimetric method of determination.² Other methods depend on diazotisation and coupling in either acid or alkaline solution. The mono-acetyl derivative melts at 219° C.

Fuller's method¹ of determining sulphanilamide has remained in use (with

modifications) in these laboratories. The urine is diluted according to the supposed sulphanilamide-content so that 100 ml. will contain about 5 mg., and to 2 ml. of the diluted liquid are added 2 ml. of *N*/2 hydrochloric acid, followed by 0.2 ml. of a freshly prepared 0.5 per cent. solution of sodium nitrite, and the mixture is allowed to stand for 3 to 5 minutes at a temperature below 20° C., and then tested with potassium iodide and starch to ensure excess of nitrous acid. If there is not an excess, more nitrite is added; otherwise 0.5 ml. of 20 per cent. urea solution is added to destroy the excess, and the mixture is left for 5 minutes. It is then treated with 0.5 ml. of a 1 per cent. solution of thymol or β -naphthol in 5 per cent. sodium hydroxide solution followed by 1 ml. of 40 per cent. sodium hydroxide solution. The resulting colour is compared with a standard or the concentration is estimated by a photo-electric colorimeter using appropriate colour filters.

Marshall, in a series of investigations,³ made use of dimethyl α -naphthylamine and acid coupling, and used ammonium sulphamate to destroy the excess of nitrous acid. Proom⁴ used the same reagent, and MacLagan⁵ introduced the photo-electric-colorimeter and the necessary colour-filters.

TABLE II

| Fuller's method | Werner's method | Remarks |
|-----------------|-----------------|-----------------------------|
| 26.0 | 47.0 | Oxidation products present. |
| 20.1 | 46.5 | Oxidation products present. |
| 107.0 | 102.0 | Oxidation products absent. |
| 125.0 | 200.0 | Oxidation products present. |
| 112.0 | 100.0 | Oxidation products absent. |
| 70.4 | 106.0 | Oxidation products present. |
| 32.0 | 42.2 | Oxidation products present. |
| 179.0 | 250.0 | Oxidation products present. |

The removal of nitrous acid is necessary, since, in presence of urine, colours are produced with the reagents, which tend to vitiate the results, although for approximate results this refinement is not essential. The purity of the dimethyl α -naphthylamine is also important, as certain oxidation products entirely alter the shade of colour; the *pH* also appears to have some effect, and in his final method Marshall buffered the solution at the same time as he removed excess of nitrous acid. His final method is as follows:—An aliquot portion of the diluted urine is treated with *N* hydrochloric acid followed by 1 ml. of a freshly-made 0.1 per cent. solution of sodium nitrite, and then, after 3 minutes, by 1 ml. of *M* sodium dihydrogen phosphate solution containing 0.5 per cent. of ammonium sulphamate. After 5 minutes 5 ml. of a 0.4 per cent. alcoholic solution of dimethyl α -naphthylamine is added and the colour is compared with a standard.

Werner⁶ condensed the amino group with *p*-dimethylamino-benzaldehyde, adding 1 ml. of a 3 per cent. solution of this reagent in 7 per cent. v/v sulphuric acid to 9 ml. of the urine previously diluted one or two hundredfold, and comparing the colour with those of similarly treated standards. Here, again, care is necessary, since the reagent itself has a pronounced colour which readily becomes

darker, and this colour may mask that produced by small amounts of sulphanilamide in the highly diluted urine.

The reagent also determines all amino groupings and so includes *p*-aminophenol, an oxidation product of sulphanilamide. Nitrous acid oxidises the phenol to quinone which does not couple. Table II compares results obtained by Fuller's diazotisation method and by Werner's method with urines in which oxidation products were present and absent. The results are expressed as mg. of sulphanilamide per 100 ml.

It has already been mentioned that the human organism acetylates the amino group before excretion, and estimations of the total sulphanilamide are necessary to compute the actual excretion of the drug, the term total being used to designate the amount of drug excreted with a free amino group plus that part in which the amino group is conjugated with acetyl radical. The acetyl radical is readily split off by heat treatment with hydrochloric acid; when, however, urine, or even diluted urine, is thus directly treated, low recovery results are obtained, but the use of nascent zinc hydroxide as a decolorising agent effects an improvement, as is shown in Table III.

TABLE III

| | Free sulphanilamide, mg. per 100 ml. | | | Acetylsulphanilamide, mg. per 100 ml. | | |
|---------------------------------|---|--------------------|-----------------------|--|--------------------|-----------------------|
| | Present Per Cent. | Found Per Cent. | Recovery Per Cent. | Present Per Cent. | Found Per Cent. | Recovery Per Cent. |
| Unheated urine decolorised | { 2 | 2.04 | 102 | | | |
| | { 4 | 3.92 | 98 | | | |
| | { 8 | 8.3 | 104 | | | |
| Heated urine diluted 25-fold | { 5 | 2.6 | 52 | 5 | 3.6 | 72 |
| | { 10 | 8.8 | 88 | 10 | 9.2 | 92 |
| Decolorised | { 5 | 4.2 | 84 | 5 | 4.8 | 96 |
| | { 10 | 10.1 | 101 | 10 | 10.6 | 106 |

In the method adopted for determining acetyl sulphanilamide the urine is heated with an equal volume of 2 *N* hydrochloric acid in a boiling water-bath for 30 minutes and then cooled, and the water lost by evaporation is replaced. Five ml. of this heated urine are treated with 2 ml. of 10 per cent. zinc sulphate solution, the *pH* is adjusted to 7-8 by addition of 10 per cent. ammonia, the whole is diluted to 10 ml., allowed to stand and filtered, and aliquot parts of the filtrate are taken. The number of dilutions must not be forgotten in making the final calculations.

Marshall's procedure differs slightly in that he heats 1 ml. of urine with 2 ml. of *N* hydrochloric acid for 30 minutes and neutralises with *N* sodium hydroxide solution before diazotisation and coupling.

For determinations in blood it is necessary first to remove proteins. In Fuller's¹ method 1 ml. of blood is laked with 2 ml. of *N*/2 hydrochloric acid, 1 ml. of 20 per cent. trichloroacetic acid is added, and the mixture is allowed to stand for a short time, and then filtered with the aid of the pump. Aliquot portions of the filtrate are diazotised, and, after removal of excess of nitrous acid, are coupled with thymol (β -naphthol must not be used, since it develops a colour with some

normal constituent of the blood filtrate). Acetyl sulphanilamide is estimated in the filtrate by heating for 30 minutes in a boiling water-bath with an equal volume of 2 *N* hydrochloric acid, cooling and diazotising.

Marshall's method³ differs somewhat, and he considers that, unless nitrous acid is destroyed, trichloroacetic acid should only be used if free sulphanilamide is to be estimated. He uses *p*-toluene sulphonic acid as a protein precipitant and his final method for blood is as follows:—One ml. of blood is laked with 7 ml. of 0.05 per cent. saponin solution, 2 ml. of 20 per cent. *p*-toluene sulphonic acid are added, and the mixture is filtered. Ten ml. of the filtrate are treated with 1 ml. of 0.1 per cent. sodium nitrite solution, followed, after 3 minutes, by 1 ml. of *M* sodium dihydrogen phosphate solution containing 0.5 per cent. of ammonium sulphamate; the liquid is allowed to stand, and then treated with 5 ml. of 0.4 per cent. alcoholic dimethyl α -naphthylamine solution, and the colour is matched. The total sulphanilamide may be estimated by heating 10 ml. of the *p*-toluene sulphonic acid filtrate in a boiling water-bath for 90 minutes, replacing the water lost by evaporation, cooling and diazotising.

The diazotisation method is suitable for the determination of those drugs in which there is a free amino group, such as Ilvin, Uleron and sulphanilamido-pyridine, or in which the amino group is readily set free as in Supron, but with the substituted sulphonamides they must be developed from the particular drug under examination, since the colours developed are often different from that produced by treatment of the correct equivalent of sulphanilamide. This is especially noticeable with sulphanilamido-pyridine, with which the colour developed is more intense than with sulphanilamide.

SULPHANILAMIDO-PYRIDINE (M. & B. 693, Dagenan).—This is a white crystalline powder, much less soluble in water than sulphanilamide and sparingly soluble in organic solvents. It melts at 191° C., and is soluble in acids and alkaline solutions, being precipitated over the range *pH* 4 to 9. The mono-acetyl derivative melts at 224° C.

Owing to its free amino group it can be estimated by direct diazotisation, but great dilution is necessary because of the intense colour produced with the usual coupling agents.

So far as I have at present been able to ascertain, neither this drug nor any other of the group in which the sulphonamide radical is substituted appears to split up to give free sulphanilamide, and there is thus little likelihood of the necessity of a "mixed estimation" arising.

Baines and Wein⁷ estimate the drug by diazotisation and coupling with dimethyl α -naphthylamine. The free amino body is estimated in blood and urine by diluting 1 ml. of the sample with 2 ml. of water and then adding 1 ml. of 20 per cent. trichloroacetic acid solution. The mixture is shaken and filtered with the aid of the pump, 1 ml. of filtrate is treated with 0.05 ml. of 0.5 per cent. sodium nitrite solution, and left for 3 minutes, after which 0.5 ml. of 1 per cent. alcoholic dimethyl α -naphthylamine solution is added and the whole is mixed and allowed to stand for 15 minutes before matching the colour with standards.

The conjugated drug is determined in urine by heating under reflux 5 ml. of the sample with 5 ml. of 20 per cent. sodium hydroxide solution for one hour,

cooling, neutralising to litmus with hydrochloric acid, and measuring the volume. To 1 ml. of this liquid, 2 ml. of water and 1 ml. of 20 per cent. trichloroacetic acid are added, and 1 ml. of this mixture is then diazotised and coupled as before; the total drug is thus determined and the amount conjugated is found by difference.

For blood the process is similar. Three ml. are treated with 1.5 ml. of water and 1.5 ml. of 20 per cent. trichloroacetic acid and the mixture is filtered. Three ml. of the filtrate are then heated under reflux for one hour with 3 ml. of 20 per cent. sodium hydroxide solution, the mixture is cooled and neutralised with hydrochloric acid to litmus, and the volume is measured. One ml. of the neutralised solution is diluted with 0.5 ml. of water and treated with 0.5 ml. of 20 per cent. trichloroacetic acid, and 1 ml. of this solution is diazotised and coupled as before.

The method I have used is similar, but involves substitution of thymol for dimethyl α -naphthylamine; the mixture is made alkaline, but decolorising with zinc hydroxide is necessary after heating under reflux with sodium hydroxide solution; for this purpose the alkaline liquid is brought to pH 8.0, 2 ml. of 10 per cent. zinc sulphate solution are added, and the whole is allowed to stand and filtered; an aliquot part of the cool filtrate is acidified with hydrochloric acid, diazotised and coupled in the usual way.

The following recoveries (Table IV) are illustrative of the results obtained.

TABLE IV

RECOVERY OF ACETYL-SULPHANILAMIDO-PYRIDINE FROM URINE

| Actual, mg. per 100 ml. | | Found, mg. per 100 ml. | | Recovery | |
|-------------------------|--------|------------------------|--------|----------------|------------------|
| Free | Acetyl | Free | Acetyl | Free Per Cent. | Acetyl Per Cent. |
| 20.0 | 4.8 | 21.0 | 3.82 | 105 | 80 |
| | 3.5 | | 3.45 | | 99 |
| | 7.5 | | 7.5 | | 100 |
| | 10.0 | | 9.7 | | 97 |
| | 6.0 | | 6.3 | | 105 |
| | 9.5 | | 9.2 | | 97 |

The drug also occurs, both free and conjugated, in vomit, etc., and the same methods of determination as for blood may be applied, but in faeces, on account of its insolubility, it becomes necessary to extract the sample by warming for 15 minutes with 10 per cent. sodium hydroxide solution; this, of course, renders it impossible to differentiate between the free and conjugated drug, although, since the conjugated form is excreted into the alimentary canal, it probably occurs in the faeces also.

PRONTOSIL AND PRONTOSIL SOLUBLE.—These two compounds have been determined by comparing the colour of the urine with that of a solution of the corresponding prontosil of known strength and of the same pH.¹ Prontosil Soluble can be estimated in presence of Prontosil by making the sample strongly alkaline, the colour due to Prontosil almost disappearing under this condition.

Both these drugs owe their activity to the liberation of sulphanilamide, and the total sulphanilamide may be estimated by boiling the strongly acidified urine

with activated charcoal (Norit), filtering, cooling, diazotising and coupling an aliquot part of the filtrate.

ILVIN.—This drug is a yellow crystalline powder (m.p. 194.5°C.), slightly soluble in water. It shows a reactive ketonic group and will form an insoluble bisulphite compound.

No examination was made for conjugated compound, although it probably occurs. The free substance may be estimated in a manner similar to that used for sulphanilamide.

It may be estimated in faeces by grinding with sand and heating under reflux with methyl alcohol. An aliquot part of the extract is mixed with dilute hydrochloric acid, diazotised and coupled.

ULERON.—This drug, in the free and conjugated form, is determined in the same manner as sulphanilamide. For the faeces, repeated extraction with $2N$ hydrochloric acid is necessary, and the extract is best decolorised by treatment with zinc sulphate.

SOLUSEPTASINE.—This is the disodium salt of *p*-(γ -phenyl propylamino) benzene sulphonamide $\alpha\gamma$ -disulphonic acid and is a white substance, readily soluble in water. It is administered by injection, and in the body some undergoes hydrolysis, liberating sulphanilamide. So far, it has not been found possible to differentiate between acetyl sulphanilamide and soluseptasine, as the heat treatment with $2N$ hydrochloric acid completely hydrolyses both compounds, but the free sulphanilamide may be estimated in the usual manner.

BENZYL SULPHANILAMIDE (Proseptasine).—This is a white, micro-crystalline powder melting at $169\text{--}174^{\circ}\text{C.}$; it is very slightly soluble in water, and more so in acid and alkali. The compound is very stable, and is hydrolysed to sulphanilamide with great difficulty. Oxidation to benzoic acid is possible, but the yield is not quantitative; nitrous acid gives an insoluble yellow product which has the characteristics of a nitroso compound, but this reaction is valueless for its determination.

TABLE V

RECOVERY OF BENZYL SULPHANILAMIDE FROM URINE

| Actual mg. per 100 ml. | Found, mg. per 100 ml. | Recovery Per Cent. |
|---------------------------|---------------------------|-----------------------|
| 6.0 | 6.3 | 105 |
| 5.9 | 6.2 | 105 |
| 1.9 | 1.9 | 100 |
| 2.2 | 2.1 | 95.5 |
| 7.5 | 7.2 | 96.0 |
| 1.5 | 2.0 | 133.5 |

On account of its stability it is still possible to estimate free and conjugated sulphanilamide by the usual methods, and the following method was devised for urine: The urine is saturated with sodium sulphate, adjusted to $pH\ 7.0$ and extracted with petroleum spirit in a continuous extractor for eight hours. The solvent is then distilled off, and the residual benzyl compound is hydrolysed by heating with 70 per cent. sulphuric acid for one hour in a boiling water-bath.

The ammonia thus produced from the benzyl compound is estimated after dilution and neutralisation.

By this procedure the results in Table V were obtained with urines in presence of free and acetyl sulphanilamide.

Continuous extraction with petroleum spirit may also be applied to dried faeces after grinding with sand.

Walti⁸ determines the compound by hydrogenation, which decomposes it into sulphanilamide and toluene; the former is then estimated by the usual method. As hydrogenation may not be convenient on account of the apparatus required, hydrolysis offers advantages.

SUPRON.—This is a compound of sulphanilamide and quinolinic acid; the sodium salt is very soluble and is given intramuscularly. In the body a small quantity of sulphanilamide is liberated and the remainder of the supron appears to be excreted unchanged.

The compound is easily hydrolysed; heating for 30 minutes in a boiling water-bath with an equal volume of 4 per cent. acetic acid effects complete hydrolysis while not affecting acetyl sulphanilamide. Both compounds are entirely decomposed by treatment with 2 *N* hydrochloric acid for 30 minutes in a boiling water-bath.

Decolorising with zinc hydroxide is necessary. By differential means it is possible to determine free sulphanilamide in addition to the other two derivatives, but the error in the amount of conjugated sulphanilamide is subject to the effect of all errors, as this substance is estimated by subtraction of the sum of the other two from total sulphanilamide.

TABLE VI

RECOVERY OF SUPRON, SULPHANILAMIDE AND ACETYL-SULPHANILAMIDE FROM URINE

| | mg. per 100 ml. | | | | | |
|--------------------|-----------------|-------|-----------------------|---------|-------|-----------------------|
| | Present | Found | Recovery Per Cent. | Present | Found | Recovery Per Cent. |
| Sulphanilamide .. | 21.5 | 25.5 | 118.5 | 10.0 | 12.4 | 124.0 |
| Supron .. | 25.0 | 22.0 | 87.0 | 40.0 | 33.3 | 83.0 |
| Total sulphonamide | 43.6 | 47.7 | 92.0 | 38.0 | 40.2 | 95.0 |
| Sulphanilamide .. | 16.0 | 14.6 | 91.5 | 16.2 | 16.8 | 104.0 |
| Supron .. | 24.0 | 25.0 | 104.0 | 16.2 | 18.2 | 112.5 |
| Total sulphonamide | 52.8 | 40.0 | 76.0 | 27.2 | 27.0 | 99.0 |
| Sulphanilamide .. | 28.0 | 31.0 | 90.0 | 33.5 | 23.0 | 68.0 |
| Supron .. | 20.0 | 23.5 | 85.0 | 24.0 | 25.0 | 104.0 |
| Total sulphonamide | 40.0 | 40.0 | 100 | 63.0 | 56.0 | 89.0 |
| Sulphanilamide .. | 16.0 | 16.6 | 104 | 20.0 | 19.2 | 96.0 |
| Supron .. | 16.0 | 16.6 | 104 | 20.0 | 20.8 | 104.0 |
| Total sulphonamide | 27.0 | 27.9 | 103 | 47.5 | 48.0 | 101.0 |

Applying the method to blood necessitated modification of the usual protein precipitant, since trichloroacetic acid on heating causes hydrolysis of the acetyl sulphanilamide, and when this acid was neutralised some hydrolysis still occurred,

as the sodium trichloroacetate becomes alkaline on heating. The use of sulphosalicylic acid as a protein precipitant solved the difficulty, and the following process was evolved:

Three ml. of blood were mixed with 6 ml. of *N*/2 hydrochloric acid and 3 ml. of 25 per cent. sulphosalicylic acid solution, shaken and filtered with the aid of the pump; aliquot parts of the filtrate were taken for the determinations of free and total sulphanilamide by the usual methods and of supron by neutralising with *N* sodium hydroxide solution to *pH* 5.0 (for 2 ml. of the filtrate approximately 0.5 ml. are required), adding 2 ml. of 4 per cent. acetic acid, heating the whole for 30 minutes on the boiling water-bath, cooling, treating with 1 ml. of *N*/2 hydrochloric acid, diazotising and coupling.

By this method the results in Table VII were obtained when different amounts of the various substances were added to normal blood.

TABLE VII
RECOVERY OF SUPRON, SULPHANILAMIDE AND ACETYL-SULPHANILAMIDE
ADDED TO BLOOD

| | mg. per 100 ml. | | | | | |
|--------------------|-----------------|-------|-----------------------|---------|-------|-----------------------|
| | Present | Found | Recovery Per Cent. | Present | Found | Recovery Per Cent. |
| Sulphanilamide .. | 4.5 | 5.2 | 116 | 3.6 | 3.6 | 100 |
| Supron | 18.0 | 20.0 | 111.5 | 7.2 | 7.2 | 100 |
| Total sulphonamide | 17.1 | 18.4 | 107 | 10.2 | 10.2 | 100 |

OXIDATION PRODUCTS OF SULPHANILAMIDE.—Many urines from patients receiving sulphanilamide darken on exposure to the air; examination of the inorganic and ethereal sulphates shows a much lower ratio than the normal 10:1. From urines of this type acetyl hydroxylamino benzene sulphonamide (1:4) and *p*-amino-phenol have been isolated, the latter especially from cyanosed patients; a purple pigment of the indophenol class may also occur, but its exact composition is not yet known.

Since hydrolysis is necessary to set these substances free, the following process has been worked out, in which the compounds are separated from the urine and also from other interfering substances.

Ten ml. of urine are heated for 30 minutes in a boiling water-bath with 0.5 ml. of conc. hydrochloric acid, and then cooled and neutralised. The neutral solution is extracted several times with ether, and the combined ethereal solutions are evaporated, water being added before the last of the ether has disappeared, since this aids the solution of the extracted compounds. The aqueous solution is made up to 20 ml. and divided into two portions for the following estimations:

***p*-Amino-phenol.**—Ten ml. of the aqueous solution are heated in a boiling water-bath for ten minutes with 2 ml. of 5 per cent. solution of phenol, then cooled, treated with 2 ml. of 2 per cent. calcium hypochlorite solution, left for ten minutes for the blue colour to develop to its maximum and compared with a standard solution of *p*-amino-phenol similarly treated. It is essential that the reagents should be freshly prepared.

Hydroxylamino benzene sulphonamide.—When pure, this is a white crystalline

powder (m.p. 161–163° C.), slightly soluble in water, its aqueous solution on exposure to air depositing fine needle crystals. For this reason standard solutions must be prepared every two days.

Ten ml. of the aqueous extract, neutralised to phenolphthalein, are treated with 10 ml. of absolute alcohol, well cooled, and treated with 2 drops of redistilled benzoyl chloride followed by 2 ml. of 2 per cent. sodium acetate solution. After thorough mixing the liquid is treated with 2 ml. of a 0.5 per cent. solution of ferric chloride hexahydrate in 2 per cent. v/v hydrochloric acid, then diluted to a known volume, and compared with a standard solution similarly treated.

It is essential that the solution after addition of the alcohol should be kept cool and that freshly prepared reagents should be used. This method, which is due to Pucher and Day,⁹ depends on the interaction of the benzoyl chloride and hydroxylamine compound to form a benzyl hydroxamic acid which produces a reddish-violet colour with ferric chloride.

Use of this method on urines, to which the compounds had been added, gave the following results (Table VIII) :

TABLE VIII

RECOVERY OF *p*-AMINO-PHENOL AND *p*-HYDROXYLAMINO BENZENE SULPHONAMIDE

| | Present Found | | | Re-covery Per Cent. | | | Present Found | | | Re-covery Per Cent. | | |
|------------------------|---------------|-----|-----|---------------------|-----|-----|---------------|-----|----|---------------------|--|--|
| <i>p</i> -Amino-phenol | 2.5 | 2.4 | 96 | 2.0 | 2.0 | 100 | 5.0 | 4.5 | 90 | | | |
| Hydroxylamine compound | 4.5 | 6.0 | 133 | 5.0 | 3.2 | 62 | 7.3 | 7.0 | 96 | | | |

A successful alternative method for the hydroxylamine compound is as follows: Add to the aqueous extract 2 ml. of *N*/2 hydrochloric acid and 1 ml. of 0.5 per cent. sodium nitrite solution, stand, add 1 ml. of 20 per cent. urea solution, again stand, then add 0.5 ml. of thymol solution and 1 ml. of 40 per cent. sodium hydroxide solution. The nitrous acid reduces the hydroxylamine compound to sulphonamide, which is determined by diazotising and coupling. Comparison should be made with hydroxylamine-compound standards which have been similarly treated.

I wish to express my thanks to Dr. L. Colebrook for performing the animal experiments and to Dr. A. T. Fuller for helpful advice.

REFERENCES

1. A. T. Fuller, *Lancet*, 1937, [i], 194.
2. F. C. Wood, *Nature*, 1937, 136, 837.
3. E. K. Marshall, K. Emerson, W. C. Cutting, *J. Amer. Med. Assoc.*, 1937, 108, 953.
E. K. Marshall, *J. Biol. Chem.*, 1937, 122, 263; *Proc. Soc. Exp., Biol. & Med.*, 1937, 36, 422. E. K. Marshall and J. T. Litchfield, *Science*, 1938, 88, 85.
4. H. Proom, *Lancet*, 1938, [i], 260.
5. E. A. MacLaglan, B. W. Carey, A. M. Butter, *J. Lab. and Clin. Med.*, 1938, 33, 1273.
6. A. E. A. Werner, *Lancet*, 1939, [i], 18.
7. E. J. Baines and R. Wein, *Quart. J. Pharm.*, 1939, 12, 4.
8. A. Walti, personal communication.
9. G. W. Pucher and H. A. Day, *J. Amer. Chem. Soc.*, 1926, 48, 672.

The Determination of Tin in Foodstuffs by means of Dithiol

By R. DE GIACOMI

THE determination of small amounts of tin in foodstuffs by gravimetric methods necessitates taking a large sample, with the attendant difficulty of the destruction of much organic matter. An attempt has therefore been made to find a suitable method which could be carried out on 10 g. of material. The colorimetric method of Schryver¹ was not found satisfactory for this purpose. On the other hand, the use of Clark's 4-methyl-1,2-dimercaptobenzene (dithiol) reagent,² with the procedure here outlined, proved successful. Hamence³ suggested the use of dithiol in a scheme designed for determining several metals in one solution. I have found, however, that the presence of ammonium sulphate, ammonium citrate and hydrogen sulphide water in the test and standard solutions renders the method unreliable.

In Clark's⁴ method agar is added to the test solution to keep the precipitate in suspension, and the colour is evaluated by means of a Lovibond tintometer. In the following method the use of agar is avoided by keeping the concentration of tin in the test solution below 30 p.p.m. The colour is matched by comparison with standard tin solutions, and this is of use when a Lovibond tintometer is not available.

REAGENTS.—(a) *Strong solution of tin*.—Tin (0.1 g.) is dissolved in 10 ml. of conc. hydrochloric acid, 0.5 ml. of thioglycollic acid is added, and the solution is diluted to 100 ml. with water.

(b) *Dilute solution of tin*.—One ml. of the strong solution of tin and 2 drops of thioglycollic acid are diluted to 100 ml. This solution should not be kept for more than one day.

(c) *Reagent*.—Dithiol (0.1g) and 0.25 ml. of thioglycollic acid are dissolved in 50 ml. of a 1 per cent. aqueous solution of sodium hydroxide.

PROCEDURE.—Careful control of the conditions under which the tin sulphide is precipitated is essential. Ten g. of the sample are treated in a 300-ml. Kjeldahl flask with 10 g. of potassium sulphate and 30 ml. of conc. sulphuric acid, and the flask is heated over a flame until the organic matter is destroyed; further additions of acid may be made if necessary. When cool, the solution is washed into a 250-ml. conical flask, so that the final volume does not exceed 100 ml. It is then neutralised with ammonia (sp.gr. 0.880), a small piece of litmus paper being used as indicator (the use of indicator solutions introduces difficulties at a later stage). Conc. hydrochloric acid is added, drop by drop, until the solution is just acid and then 2 ml. excess. The litmus paper is rinsed and removed, the solution is cooled, and hydrogen sulphide passed through it first for 10 minutes in the ordinary way and then for 1 hour under pressure, obtained by passing the delivery tube through a rubber stopper inserted firmly in the neck of the flask. The solution is next boiled gently for 5 minutes and filtered through a Buchner funnel containing a No. 5 Whatman filter-paper, 4.25 cm. in diameter. The filtrate is re-filtered two or three times until it is clear and colourless, after which it is again saturated with hydrogen

sulphide, allowed to stand under pressure for half-an-hour, again boiled, and filtered through the original filter. If the filtrate is not crystal clear the filtration is repeated, but this is not usually necessary. The flask and filter are washed two or three times with water. The filter-paper is placed in a squat 100-ml. beaker, 10 ml. of 10 per cent. sodium hydroxide solution are added, and the beaker is heated on a water-bath for at least 10 minutes. The pulp and solution are poured on to the funnel previously used, a No. 5 paper again being used, and the beaker and paper are washed several times by decantation with water (10 to 20 ml. in all). The filtrate is washed into a 100-ml. flask and made just acid by adding conc. hydrochloric acid, drop by drop, a small piece of litmus paper being used as indicator. Two drops of thioglycollic acid are added, and the solution is made up to 100 ml. with water.

Five ml. of the solution are treated in a boiling-tube with 5 ml. of water, 0.5 ml. of conc. hydrochloric acid and 0.5 ml. of reagent. The tube is immersed in a boiling water-bath for 30 seconds and then allowed to stand for 1 minute, after which the colour is compared with standards prepared in the same way from the dilute tin solution and made up to the same volume. The comparison is made by viewing the tubes from the side by light reflected at right angles. When an approximate match has been obtained the test is repeated, the standard and test solutions being prepared simultaneously. To obtain the best results the amount of test solution taken should not give a greater depth of colour than is given by 4 ml. of the dilute tin solution. The test solution is diluted accordingly, or smaller aliquot portions are taken, to bring it within this range. If tin is present in a very small quantity, 10 ml. of the test solution are used.

TABLE I

| Expt. | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 |
|---------------|------|------|------|------|------|------|------|------|------|------|------|------|
| Added, mg. | 0.30 | 0.70 | 0.75 | 1.00 | 1.25 | 1.40 | 1.50 | 1.75 | 2.10 | 2.80 | 4.00 | 6.00 |
| " p.p.m. | 30 | 70 | 75 | 100 | 125 | 140 | 150 | 175 | 210 | 280 | 400 | 600 |
| Found, mg. | 0.25 | 0.65 | 0.75 | 0.90 | 1.20 | 1.30 | 1.40 | 1.60 | 2.00 | 2.60 | 3.75 | 5.20 |
| " p.p.m. | 25 | 65 | 75 | 90 | 120 | 130 | 140 | 160 | 200 | 260 | 375 | 520 |
| Diff., p.p.m. | 5 | 5 | — | 10 | 5 | 10 | 10 | 15 | 10 | 20 | 25 | 80 |

The experimental results given in Table I were obtained by adding known amounts of standard tin solution to 10 g. of minced beef or veal known to be free from tin.

It will be observed that above 150 p.p.m. the error increases. This is necessarily a limiting factor in all colorimetric work. The error was found to be chiefly due to small differences in the tint between the standard and test solutions; by adopting the following modification greater accuracy may be achieved with the higher concentrations of tin.

The dilute solution of tin is prepared by adding to 1 ml. of the strong solution of tin 95 ml. of water and 5 ml. of conc. sulphuric acid. The solution is then treated in exactly the same way as the acid solution obtained after the destruction of the organic matter. The dissolved sulphide is treated with hydrochloric acid and thioglycollic acid as before, and made up to 100 ml., and this solution is used as the standard. Some results thus obtained are given in Table II.

TABLE II

| Expt. | | Mgms. | p.p.m. | Diff. p.p.m. |
|-------|-------|-------|--------|--------------|
| (a) | Added | 1.50 | 150 | nil |
| | Found | 1.50 | 150 | |
| (b) | Added | 2.00 | 200 | nil |
| | Found | 2.00 | 200 | |
| (c) | Added | 3.00 | 300 | nil |
| | Found | 3.00 | 300 | |
| (d) | Added | 4.00 | 400 | 15 |
| | Found | 3.85 | 385 | |

OBSERVATIONS.—(a) In the destruction of organic matter the use of nitric acid is avoided, since this may give rise to a precipitate of insoluble metastannic acid, which has to be filtered off and fused with caustic soda.

(b) The reagent is best kept in an atmosphere of hydrogen and should be rejected on the appearance of a white precipitate of the disulphide. I have been unable to preserve the solution satisfactorily for more than two weeks.

(c) If the filter is washed by decantation, not by means of a jet, there is no risk of colloidal tin sulphide being carried through it. By using No. 5 Whatman paper, in conjunction with a Buchner funnel and a suction pump, a fine filter, which retains the precipitate, is obtained.

(d) Specimen tubes (6 in. \times 1 in.) have been found more suitable than boiling-tubes for comparing the solutions. They should be selected so as to be of the same diameter and of the same tint of glass.

(e) The colour matching should be carried out by daylight, as I have found that with artificial light accurate work is impossible. Also, the matching should be carried out on the same day as that on which the final test solution is prepared.

REFERENCES

1. G. S. Buchanan and S. B. Schryver, *Local Govt. Board Report*, No. 7, 1908.
2. R. E. D. Clark, *ANALYST*, 1936, **61**, 242.
3. J. H. Hamence, *id.*, 1937, **62**, 18.
4. R. E. D. Clark, *id.*, 1937, **62**, 661.

November 9th, 1939

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

AN ALKALINE AGEING TEST FOR TEXTILE OILS

OILS for lubricating textile fibres should be examined for liability to undesirable oxidation, from two aspects:—(a) The rate of oxidation (which depends upon the catalyst/antioxidant balance and is measured by such methods as the Mackey test and the peroxide accumulation rate); (b) the type of end-product (which depends upon the chemical constitution and is indicated by the yield of petrol-insoluble bromides, iodine value of the liquid fatty acids, etc.).

The present test gives visual evidence of the formation of undesirable end-products, by the production, under controlled laboratory conditions, of effects similar to those that may result from the use, in bulk, of unsuitable oils. White worsted serge, previously extracted with ether to remove soap and oil, is cut into 3-inch squares; 0.1 ml. of the oil is dropped into the centre of one of these squares, which is then suspended in an oven at 80° C. six inches above a dish of dilute ammonia (100 ml.). After 4 hours the pattern is removed from the stove and cut into two halves. One-half is retained for reference; the other is scoured for 10 minutes in 300 ml. of a solution containing per litre 0.2 per cent. of soap and 0.1 per cent. of ammonia at 40° C., drained, scoured again in the same solution, washed off, dyed in a dye bath (5 g. of serge) containing 0.04 g. of Coomassie Blue R.L.S. (I.C.I.), 10 g. of Glauber's salt and 3 ml. of acetic acid (added in portions of 1 ml. at 20-minute intervals), beginning cold, heating to boiling during 20 minutes, and boiling for 1 hour.

The following table gives some typical results:

| Oil | Discoloration after alkaline ageing | Resist after scouring and dyeing | Mackey test (mins. to 400° F.) |
|------------------------------------|-------------------------------------|----------------------------------|--------------------------------|
| Nilox ester oil | Brownish | nil | No rise |
| Nilox arachis oil | Cream | nil | No rise |
| Olive oil | Pale yellow | Very slight | 215 |
| Olive oil containing iron | Pale fawn | Slight | 125 |
| Arachis oil (edible) | Fawn | Slight to mod. | 310 |
| Cottonseed oil (edible) | Brownish-fawn | Moderate | 245 |
| Linseed oil (refined) | Brownish-yellow | Great | 95 |
| Linseed oil containing antioxidant | Pale brownish-yellow | Great | 340 |

The discoloration produced by the alkaline ageing appears to be due partly to oxy-acids (which form brownish alkali salts) and partly to yellowish dimers and polymers resulting from the oxidation and condensation of constituents which yield petrol-insoluble bromides.

The depth of colour increases with the unsaturation (*e.g.* linseed oil becomes darker than olive oil), the amount of oxy-acids present (*e.g.* blown olive oil becomes darker than olive oil), and the amount of effective catalyst (*e.g.* olive oil with a bad Mackey test and high peroxide accumulation rate becomes darker than an olive oil with a slow rate of oxidation).

The coloured bodies due to oxy-acids are scoured out easily and do not cause a resist in dyeing; those due to the condensation products are not scoured out, and the residue after scouring acts as a resist in dyeing. It is the resist effect after dyeing which is therefore to be taken as the significant indication of unsuitability.

It will be noticed that the test is sensitive to the presence of effective catalysts (which are not shown by such tests as the yield of petrol-insoluble bromide) and of di- or poly-ethenoids (which are not shown by the Mackey test, etc.); the effect of polyethenoids is only slightly masked by the presence of antioxidants.

An oil which produces a resist greater than that given by an olive oil that is free from antioxidant and has a good Mackey test and low peroxide accumulation rate should be considered unsuitable for textile use.

An oil which gives a resist less pronounced than this standard is not necessarily suitable, but if, in addition, it answers to the following requirements it can be considered reasonably safe:—iron-content less than 1 p.p.m., Mackey test and peroxide accumulation rate satisfactory, and yield of petrol-insoluble bromides less than 10 per cent. (octo- and hexa-bromides being absent). W. GARNER

March, 1940

UNIVERSAL BUFFER MIXTURE

IN THE ANALYST, 1939, **64**, 490, Johnson and Lindsey described what they regard as an "Improved Universal Buffer Mixture," this being presumably an improvement on the mixture introduced by Robinson and myself in 1931 (*J. Chem. Soc.*, 1931, 1456). Apparently they have overlooked the fact that Welford and I described the same modification in the *J. Chem. Soc.*, 1937, 1848, and, moreover, calibrated it for the temperatures: 12·5°, 25°, 34° 53', 63°, 75°, and 91° C., instead of at one temperature, viz. 18° C., as Johnson and Lindsey have done. This Universal Buffer mixture has the advantage over other similar mixtures, that none of its acids is volatile, so that it can therefore be used for *pH* work at elevated temperatures. H. T. S. BRITTON

March, 1940

COPPER IN TOMATO PULP

IN the notice of the Annual Report of the Chief Medical Officer of the Ministry of Health (ANALYST, 1940, **65**, 103) reference is made to an agreement reached at a conference of Port Medical Officers of Health in October, 1938, as to the tolerance of copper in imported tomato pulp.

Information has now been received that Port Medical Officers of Health, at a later conference, agreed to postpone until further notice the intended reduction of the tolerance from 100 to 50 parts per million in the dried total solids.

EDITOR

Notes from the Reports of Public Analysts

The Editor would be glad to receive the Annual or other Reports of Public Analysts containing matter of special interest to the Society. Notes made from such Reports are submitted to the Publication Committee.

COUNTY OF KENT

REPORT OF THE COUNTY ANALYST FOR THE FOURTH QUARTER, 1939

OF the 694 samples received from the County sampling officers, 43 were purchased informally.

TOWN REFUSE AS MANURE.—Two samples of treated household refuse from different sources gave the following percentage results:

| Water | Nitrogen | Phosphoric acid | Potash | Organic matter |
|-------|----------|-----------------|--------|----------------|
| 7.7 | 0.67 | 0.59 | 0.24 | 33.1 |
| 6.6 | 0.80 | 0.51 | 0.22 | 30.8 |

In both samples the organic matter consisted essentially of paper and carbon in the form of partly burned coal. Household refuse is often advocated as an organic manure, but the proportion of organic matter, even in the dried refuse, is invariably comparatively low. In these two samples only about one-third of the material was organic in nature and the preponderating substance was coal ash, ash material amounting to more than 60 per cent. The value of treated house refuse can only be ascertained by actual experiment, and at the moment these experiments would best be carried out on the soils to which the manure might be applied in the future. The results of a few experiments carried out at both Rothamsted and Woburn have been published (*cf.* ANALYST, 1940, 104), and they would tend to show that a proportion, at all events, of the nitrogen in house waste is in a form that renders it available as a plant food. It may well be that these treated household refuses would have a manurial effect extending over two years and, further, they may advantageously affect the soil condition. It seems to me that whether these wastes will ever attain popularity must depend on the price at which they can be brought to the farm. Rather large quantities will be necessary to supply the available nitrogen essential for crop production, and still larger quantities if dressings are required to improve soil tilth. F. W. F. ARNAUD

Legal Notes

Under this heading will be published notes on cases in which points of special legal or chemical interest arise. The Editor would be glad to receive particulars of such cases.

IS CARBOLIC ACID A DRUG?

ON February 3rd a druggist was summoned at Bishop Auckland under the Food and Drugs Act for selling carbolic acid not of the quality demanded.

Mr. H. C. Underwood, prosecuting for Durham County Council, said that two bottles of carbolic acid had been purchased from the defendant by an inspector under the Food and Drugs Act. The substance was labelled "Carbolic Acid (Crude). Contains Phenols 90 per cent. weight in volume. For disinfecting purposes." Whilst according to the label it contained 90 per cent. of phenols, analysis showed it to contain only 14 per cent. Although there was no fixed

standard for crude carbolic acid, the B.P. stated that carbolic acid contained not less than 98 per cent. of phenols. The fact that the article was sold for use as a disinfectant brought it within the meaning of a drug.

Mr. C. J. H. Stock, F.I.C., County Analyst, said that the sample examined by him contained only 14 per cent. of phenols. In his opinion, such a substance was practically valueless as a disinfectant. In cross-examination he said that he thought that there was some doubt whether the substance came within the Food and Drugs Act.

Mr. Brown-Hughes, for the defence, contended that the product could not be described as a medicine, and, if that were so, it could not be defined as a drug coming within the ambit of the Food and Drugs Act. He submitted that he had no case to answer.

Mr. Underwood maintained that the term drug meant anything that could be used in the prevention or alleviation of disease, and, as a disinfectant came within this category, it should therefore come within the scope of the Food and Drugs Act.

The Magistrates decided that there was no case to answer and dismissed the charge.

CARBOLIC OINTMENT

ON February 26th a druggist was summoned at Stalybridge for selling carbolic ointment deficient in carbolic acid to the extent of 92 per cent.

Mr. Gregory, prosecuting for the Cheshire County Council, said that, as there was no fixed standard for carbolic ointment, the magistrates could fix their own standard on the evidence.

Mr. S. E. Melling, F.I.C., Public Analyst for Cheshire, said that the sample of ointment examined by him contained 0.24 per cent. of carbolic acid. The British Pharmacopoeia did not fix a standard for the ointment, but prescribed that, in compounding it, 3 per cent. by weight of phenol should be added; in his opinion, as the sample contained less than one-tenth of 3 per cent., it was of very little use as an antiseptic.

In cross-examination the witness said that phenol might evaporate to the extent of 28 per cent. unless kept in an air-tight container. The British Pharmacopoeia did not give directions as to keeping carbolic acid, and witness agreed that ointment kept in the jar produced in court might lose 25 per cent. of its phenol by evaporation in 2 years.

Mr. Henriques, for the defence, said that before the magistrates could fix a standard there must be evidence before the Court from which they could assume a standard recognised in the trade.

Dr. Mumford, F.R.C.P., consulting dermatologist at the Manchester Royal Infirmary, said that the medical profession was not in the habit of prescribing carbolic ointment. For medicinal purposes 0.24 per cent. of phenol in carbolic ointment would be as effective as 3 per cent.; in other words, it would be "completely ineffective." Three per cent. of phenol might be dangerous if used on an open cut or abrasion. The only form in which he prescribed carbolic acid therapeutically was as a 1 per cent. lotion.

The defendant said that he had obtained the ointment from a firm of manufacturing chemists and had kept the jar on a shelf in his shop. There was very little demand for it, and he had not sold more than 2 oz. a year.

Mr. Thomas Tickle, F.I.C., said that the sample he had analysed for the defendant contained 0.36 per cent. of phenol. There was no acknowledged standard; knowing what he did of the sample, he would regard it as genuine.

The Magistrates decided that the case for the prosecution had not been proved, but would not allow costs.

Department of Scientific and Industrial Research

METHODS FOR THE DETECTION OF TOXIC GASES IN INDUSTRY

CARBON MONOXIDE*

OCCURRENCE.—Among the industrial situations in which this gas may be encountered in dangerous concentrations are blast furnaces, brick kilns, chemical works, foundries, gas works, coke ovens, lime kilns, and gas and water gas producers.

POISONOUS EFFECTS.—The first symptoms of poisoning are shortness of breath and palpitation on exertion, accompanied by headache which increases in severity. With increasing intoxication the judgment becomes disturbed, and the affected person may not realise his danger. An atmosphere in which a concentration as low as 1 part in 2000 is present may prove fatal in about one hour to a person engaged in an active occupation. On the other hand, concentrations below 1 in 10,000 may be regarded as relatively harmless for all practical purposes.

METHODS OF DETECTION.—Methods based on the reaction of carbon monoxide with haemoglobin, though accurate and sensitive, are unsuitable for industrial purposes. The iodine pentoxide method has the drawback that the apparatus and the procedure are too complicated for routine tests.

The standard method developed consists in drawing samples of the atmosphere under examination through a known area of test-paper impregnated with palladium chloride, at a slow and constant rate, by means of a 5-litre aspirator.

The palladium chloride solution is prepared by dissolving 0.1 g. of the pure salt in about 20 ml. of boiling water, filtering and cooling the solution, and making up the filtrate to 20 ml. This solution is then mixed with 20 ml. of pure acetone. The test-papers (3 in. × 2 in.) are impregnated with this solution.

To remove interfering gases the sample of air is made to pass through a tube of activated charcoal before passing through the test-paper. Sampling is continued until a stain is obtained on the test-paper which comes within the range of the standard colour chart; the concentration is then found by comparing the time required to reach the necessary colour with the times given on the chart. In this way concentrations of 1 part in 500 can be detected in less than two minutes, and of 1 part in 10,000 in half-an-hour. Full instructions for carrying out the test are contained in the leaflet.

In the colour chart, which is included in the Report, two standard stains are given, and it is directed that the test-paper should be examined at intervals of 5 minutes until a stain is obtained which is darker than Standard No. 1 and lighter than Standard No. 2.

FOOD INVESTIGATION BOARD

REPORT FOR THE YEAR 1938†

THE most important development during the year under review has been the expansion of the work of the Board to cover research on the processing of food. A committee has been set up to consider the organisation of research in this field in the interests alike of the consumer, producer and manufacturer. Although some work has already been carried out on the manufacture of bacon, the smoking and salting of fish and the canning of fruit, the work of the Board has previously

* Leaflet No. 7. H.M. Stationery Office, York House, Kingsway, London, W.C.2. 1939. Price 1s. 6d. net.

† H.M. Stationery Office, York House, Kingsway, January, 1940. Price 4s. 6d. net.

been mainly concentrated on the problems involved in preserving the "fresh" properties of unprocessed food during transport and storage.

TRANSPORT OF MEAT.—Attention is directed to the progress in the transport of chilled beef from Australia and New Zealand by gas storage in an atmosphere enriched, to a controlled extent, with carbon dioxide. During the five years since the method was introduced our imports of chilled beef from Australia and New Zealand have increased nearly tenfold to a combined annual figure of 850,000 cwts. Experiments have indicated that the "bloom" of the meat might be improved by increasing the rate of evaporation from the meat in the course of the voyage.

WASTAGE OF ORANGES IN TRANSPORT.—During the year arrangements were made to carry out on behalf of the Government of Palestine a survey of the wastage of oranges during transport. The temperature in the holds is an important feature in the wastage of the fruit, and the main object was to ascertain how far temperature variations were responsible for the wastage and to what extent the temperature conditions might be improved. Experiments were also carried out on the storage conditions of Palestinian grapefruit after its arrival in England.

GAS STORAGE OF FRUIT AND VEGETABLES.—Further successful experiments have been carried out on the gas storage of home-grown pears, apples and broccoli. No form of storage for apples can be reckoned successful unless it conserves their flavour, and this is especially important in the case of Cox's Orange Pippin. A full-scale demonstration was arranged to remove any doubt in the trade as to whether this variety developed its full flavour after gas storage. Twenty-six tons of Cox's Orange Pippins were put into a gas store at the Ditton Laboratory at the end of September, 1937. The composition of the atmosphere of the store was 2.5 per cent. of oxygen, 5 per cent. of carbon dioxide and 92.5 per cent. of nitrogen, and was obtained by the removal of the excess of carbon dioxide by a scrubber of commercial design, together with controlled ventilation. The temperature of storage was 39° F. The store was opened on February 22nd, 1938, in the presence of about 150 fruit-growers and other experts. The demonstration was completely successful, the fruit being in excellent condition; in fact, 80 per cent. of it was graded as "Fancy" or "Extra Fancy." On the other hand, a subsequent survey of fruit of this variety from 10 representative areas has shown that the extent to which the flavour is developed depends, in the main, on pre-storage conditions, such as soil, manurial treatment and maturity at the time of gathering.

GLYCOGEN IN MEAT.—No adequate explanation of the variations in the glycogen-content of pig's muscle is yet forthcoming. The amount of glycogen in the livers of oxen is usually about 1 per cent., as compared with about 0.1 per cent. in pig's liver. Animals whose muscle contains little glycogen also have little in the liver, so that the deficiency is general and not confined to the muscular tissue.

RANCIDITY OF HERRING OIL.—The potency of the enzyme that causes oxidative rancidity in herring oil is decreased by storing the fish at temperatures ranging from 0° to -10° C., the decrease being most rapid at the higher temperatures.

ESTIMATION OF THE FRESHNESS OF FISH.—Rapid chemical methods for determining the concentration of di- and tri-methylamines have been devised. The determination of either compound can be used as a means of detecting incipient staleness, but to follow the course of deterioration prior to this stage it is necessary to make a series of colorimetric estimations of dimethylamine.

"PINK" SPOILAGE OF SALTED FISH.—The micro-organisms responsible for this type of spoilage have been found to belong to two groups, *Serratia* and *Micrococci*, the latter being responsible for the final stage of deterioration. The best remedy, so far discovered, is to chill the fish below 5° C.

METABOLISM OF POTATO CARBOHYDRATES.—The potato tuber has been found to contain a zymohexase, and by the action of this enzyme on hexosediphosphate

(the presence of which has also been established) triosephosphate is formed; this transformation is regarded as the initial stage of the breakdown of sugar in yeast and in muscle.

STIMULATION OF RIPENING OF PLUMS BY ACETYLENE.—The recent finding in South Africa (*Nature*, 1938, 141, 876) has been confirmed. Immature *Monarch* plums, when treated at 18.3° C. with an atmosphere containing acetylene, ripened more rapidly and developed a better flavour than untreated plums.

ENGINEERING.—Among the other investigations described is the effect of turbulence in increasing the rate of evaporation from a wet surface. Further studies have also been made of the properties of refrigerants.

All-India Institute of Hygiene and Public Health

ANNUAL REPORT FOR THE YEAR 1938

IN discussing the work of the Institute (*cf.* ANALYST, 1939, 64, 196) the Director (Dr. John B. Grant) directs attention to the paradox that, whilst it is fully recognised that the country is badly in need of a vigorous and forward policy with regard to public health organisation, even the small number of trained workers are unable to find employment; this is due to the magnitude of the problem on the one hand and to the shortness of funds on the other.

In addition to the teaching work various investigations have been continued. These include the following:

SALMONELLA ENQUIRY.—Fever and diarrhoea of unknown origin are widely prevalent in Indian towns. Preliminary investigations have shown that a high percentage of rats, caught at random in Calcutta, carry salmonella organisms. The facts so far revealed should serve to stimulate public interest in the destruction of this pest, even in the absence of plague.

STUDIES IN CALCIUM METABOLISM.—Three types of Indian diets: (1) good North Indian, (2) well-to-do lacto-vegetarian, and (3) poor Indian, were fed to groups of rats. Each group was made to bear its litter to enable the study to be continued over three generations, and, for comparison, a control group was fed on a well-balanced diet. The bones and teeth were studied by four methods: (1) X-ray pictures, (2) chemical analysis of bone and teeth, (3) histological, and (4) micro-incineration of histological sections. X-ray pictures showed lighter shadows as calcium intake was decreased, and chemical analysis showed that the percentage of ash from the bones decreased with lower calcium intakes. In the teeth, however, the percentage of ash remained fairly constant, and the calcium: phosphate ratio remained practically constant in both the bones and teeth in all groups. Histological examination revealed no difference in the bones, but striking differences were noticed in the structure of the teeth.

Micro-incineration of histological sections and examination of the ash under dark-ground illumination is an excellent method for the study of inorganic constituents in cells.

One conclusion drawn from the investigation is that even the North Indian diet, which is considered to be ideal, is inferior to the stock diet if the histological picture of the teeth is studied.

VITAMIN A CONTENT OF FISH OILS.—Oils obtained from 15 varieties of local fresh-water fish were examined. The body and depot oils were very poor sources of vitamin A, but the liver oils contained more than was present in an ordinary brand of Norwegian cod-liver oil. The liver oils of two species, "Air" and "Boal,"

had a vitamin A content nearly half that of halibut liver oil, and the liver oils of "Dhain" and "Shole" were nearly equal to halibut oil. It would thus appear that the industrial preparation of Bengal fish-liver oils would be feasible. These oils showed the typical absorption bands of vitamin A₂.

DETECTION OF SULPHANILAMIDE POISONING.—Since a certain number of patients using sulphanilamide preparations have developed severe types of anaemia, the popularity of these drugs has greatly suffered. The anaemia is due to the formation of sulphaemoglobin in the blood, and this substance can easily be detected by the spectrograph, as it gives specific absorption at 6200 Å. Blood samples from a group of patients receiving diaminodiphenyl sulphone glucoside were examined daily, and it was noticed that sulphaemoglobin could be detected about six days after the beginning of the treatment and a week before any clinical signs of anaemia could be observed. It would thus appear that a spectrographic examination of blood will be a useful guide in chemotherapy with sulphanilamide derivatives. It is interesting to record that in meningitis patients large doses of the drug were administered without the appearance of sulphaemoglobin in the blood.

EPIDEMIC DROPSY AND ARGEMONE MEXICANA OIL.—In the previous Report (ANALYST, 1939, 64, 196) it was shown that the use of certain kinds of mustard oil produced epidemic dropsy. It was found that the incriminated oils gave a brown colour in Hauchecorne's nitric acid test and a green colour when mixed with an equal volume of glacial acetic acid and treated with a 3 per cent. solution of cupric acetate. Moreover, the toxic oils had a characteristic bluish-violet fluorescence in ultra-violet light, and, when examined spectrophotometrically, showed a broad absorption band between 2900 Å and 2600 Å with a maximum about 2750 Å. The possibility of the mustard seeds having been mixed with some poisonous seeds was investigated, and it was found that the oil expressed from a mixture of mustard seed with 6 per cent. of *Argemone mexicana* seeds answered to the same physical and chemical tests as the known toxic oil from Rangpur. A white crystalline substance, soluble in hot alcohol and in dilute hydrochloric acid, was isolated from *Argemone* oil, and this gave intense reactions in the differential tests. Further work on this substance is in progress. *Argemone mexicana* grows wild and is plentiful in different parts of India, and the seeds may get accidentally mixed with mustard seed at the time of harvesting or possibly there may be intentional adulteration before the seeds reach the millers.

Commonwealth of Massachusetts

ANNUAL REPORT OF THE DIVISION OF FOOD AND DRUGS DEPARTMENT OF PUBLIC HEALTH

THE Report of the Director of the Division (Dr. H. C. Lythgoe) for the year ending November 30th, 1938, gives an outline of the routine work relative to the enforcement of the laws pertaining to the sale of milk, sale of food and drugs, the bakery law, mattress law, etc. The total number of milks examined was 5154, of which 529 were below standard. Of the 2325 samples of other foods examined, 417 were adulterated, and 18 of the 182 samples of drugs were adulterated or misbranded. Among the points of interest discussed are the following:

GOATS' MILK.—A New York dairy is now selling pasteurised certified goats' milk in Massachusetts, and one goat breeder in the Commonwealth sells the milk pasteurised. Exaggerated claims are made as to the curative and medicinal properties of this milk, and there is some risk of its becoming a fad. Investigations have shown that practically all the animals are tested for Bang's disease as well

as for tuberculosis; recently one animal reacted to the tuberculin test. The following table summarises the results of the analysis of samples of milk of known purity obtained from 21 goats:

| | | | | Highest Per Cent. | Lowest Per Cent. | Average Per Cent. |
|------------------------------------|----|----|----|----------------------|---------------------|----------------------|
| Total solids | .. | .. | .. | 18.40 | 11.96 | 14.42 |
| Fat .. | .. | .. | .. | 7.25 | 2.95 | 4.93 |
| Solids-not-fat | .. | .. | .. | 11.17 | 8.39 | 9.49 |
| Lactose .. | .. | .. | .. | 5.65 | 4.25 | 4.78 |
| Total proteins | .. | .. | .. | 5.62 | 3.38 | 4.11 |
| Albumin .. | .. | .. | .. | 1.37 | 0.77 | 1.06 |
| Ash .. | .. | .. | .. | 1.04 | 0.75 | 0.89 |
| Copper serum refraction at 20° C. | | | | 40.2 | 36.3 | 38.2 |
| Acetic serum ash, g. per 100 ml... | | | | 1.105 | 0.845 | 0.970 |
| Freezing-point, °C. | .. | .. | .. | -0.550 | -0.595 | -0.573 |
| Protein: fat ratio .. | .. | .. | .. | 0.67 | 1.30 | 0.83 |
| Albumin in proteins, per cent... | | | | — | — | 25.79 |

The average total solids are somewhat higher than for cows' milk, as is also the fat; the total proteins are considerably higher, as is also the ash. Hence, the average freezing-point applied to cows' milk for the detection of added water is too high for goats' milk.

ADDED WATER IN SHELLFISH.—Of 145 samples of shelled clams examined, 85 were found to have been soaked, *i.e.* allowed to absorb a considerable amount of water. Prosecutions were instituted in many of these cases. Three samples of oysters containing added water were also obtained.

PORK IN HAMBURG STEAK.—A sample of Hamburg steak containing pork was taken in Western Massachusetts. The store manager admitted having used up pork ends in this way, and stearine crystals characteristic of pork fat were separated from the fat of the sample. It is unfortunate that pork is so often put into Hamburg steak, because the material is assumed by the public to be beef. It is frequently eaten semi-raw and is very rarely cooked at a temperature sufficiently high to kill trichinac. It is possible that the apparent increase in trichinosis in this part of the country may be due to the practice of adding pork to Hamburg steak without giving a notification to the customer.

COLON BACILLI IN CRAB AND LOBSTER MEAT.—The bacterial counts of crab and lobster meat indicated that the business is not conducted under any greater degree of sanitation than in the previous year, when so much sickness resulted. Of 18 samples of crab meat examined, 14 contained colon bacilli with counts ranging from 220 to 95,000. All the 10 samples of lobster meat contained colon bacilli with counts ranging from 260 to 70,000.

COLON BACILLI IN VEGETABLES.—During September, 1938, there was a disastrous flood in Western Massachusetts. Eleven samples of vegetables which had been in the flood were examined and 5 contained *B. coli*. Carrots that had been in the flood waters were found, after the usual washing in cold water, to be free from colon bacilli, but cabbages were found to have been contaminated.

Manchester Chamber of Commerce

TESTING HOUSE AND LABORATORY

ANNUAL REPORT FOR THE YEAR 1939

THE outbreak of war caused a decrease in the work of the Testing House, but the position is rapidly becoming normal and large orders placed for Empire defence purposes are being inspected. A special feature has been the inspection, for local A.R.P. authorities, of gas-proof oiled fabric suits. Investigations of general interest include the following:

STAINS AND DEFECTS IN FABRICS.—Bleaching of the blue "silk" trimming of a masonic apron was found to be due to its contact with the goatskin, which was strongly acid; the cotton weft in the ribbon had also suffered loss in strength.

Pink Staining of Bleached Cotton.—In one instance this was due to aniline vapour, but in another it set an interesting problem. Pink stains developed on stocks of white polo helmets, and these could only be attributed to the vapour from amino-compounds in the rubber solution used for securing the bleached drill to the cork foundation.

Red stains on coloured frocks were attributed to a perspiration corrective affecting the dyestuffs.

SURGICAL DRESSINGS.—The total number of dressings submitted under the National Health Insurance Acts was 1156, and 69 of these (6 per cent.) were deficient in quality and 20 (1·7 per cent.) in quantity. The corresponding figures for the previous year were 5·8 and 1·3 per cent. Dressings supplied in the original sealed packets showed less deficiencies than those not so packed. A number of samples of white and boric lint contained fewer weft threads than the standard. A more serious defect was deficient absorbency, some of the dressings taking several hours, instead of 10 seconds, to sink in water. Other materials, mainly bandages, yielded two or three times the permissible amount of foreign matter. The proportion of medicament in a number of dressings differed considerably from that specified. Thus a sample of boric lint contained 55 per cent. of boric acid instead of 35 to 45 per cent.; a double cyanide gauze had a total cyanide-content of 1 per cent., instead of the 2 to 4·5 per cent. required, and an iodoform gauze contained no perceptible amount of iodoform, whereas 4 to 6 per cent. was stipulated.

Adhesive-coated or rubber- or oil-proofed appliances were deficient in adhesive or proofing, some to the extent of 40 per cent., and the fabrics used were sometimes low in weight by as much as 20 per cent. and contained fewer threads than required.

COTTON WOOL.—Many samples were inferior to the standard in respect of freedom from "neps" and particles of leaf and shell. One sample contained pieces of coloured thread in addition to particles of cotton seed, and hard bast fibres were found in another sample. A third sample contained 25 per cent. of rayon fibre, presumably added to impart a soft "handle" to the material.

South Africa

ANNUAL REPORT OF THE DIVISION OF CHEMICAL SERVICES, 1939

THE Division of Chemical Services (Chief: Dr. J. P. van Zyl) undertakes work for all the State Departments and for provincial administrations. Its work consists of (a) regulatory and control work, and (b) investigational and research work. The chemical work required in the working of Acts administered by the different Departments comes under the first category, whilst the second category comprises work on soil classification, soil fertility and micro-biological problems, food and nutrition, toxicological research and miscellaneous agricultural and industrial investigations.

REGISTRATION OF FERTILISERS AND FEEDING STUFFS.—All applications are scrutinised by the Division before a list of fertilisers and farm foods registered with the Department of Agriculture and Forestry is published. This list gives the registered number, the name of the person registering, and the chemical composition of fertiliser or farm food, so that the public can see exactly what products are on the market and an official check can be kept to see that they conform to their declared composition.

ARSENIC IN PEARS AND APPLES.—The total number of samples examined was 4202, and 430 were found to contain arsenic in excess of the legal export limit of 1/100 grain of arsenious oxide per lb.

FOOD AND DRUGS.—Of the 5235 samples examined for the Department of Public Health, 545 were adulterated or below standard. The quality of milk was still unsatisfactory, for 321 of the 3435 samples examined were adulterated or below standard. Of 223 samples of ice-cream received, 40 contained less than the 10 per cent. of fat required by the regulations.

EFFECT OF FLUORINE INSECTICIDES ON CITRUS TREES.—Experiments with fluorine insecticides, such as cryolite and sodium fluosilicate, have been continued, to ascertain if there is any danger involved in the use of these remedies in the form of spray or dust against American ball worm. The young oranges are treated when they are about the size of a pea, and when ripe both the fruit and peel are analysed for fluorine. This work has already been in progress for 3 years, in view of the possibility that the effect may be cumulative. The effect of superphosphates has also been tested, since most superphosphates contain about 1.5 per cent. of fluorine. The results so far obtained indicate that there is no danger of navel oranges taking up fluorine from the soil or of the quality of the oranges being affected by dusting or spraying with fluorine insecticides.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Determination of Creatinine with *m*-Dinitrobenzoic Acid. E. Komm and H. Pinder. (*Z. Unters. Lebensmittel*, 1939, 78, 113–123.)—The suitability of *m*-dinitrobenzoic acid for the quantitative determination of creatinine has been confirmed; it is more specific than picric acid. A disadvantage of picric acid is its own colour, whereas *m*-dinitrobenzoic acid is only pale yellow, and after purification is quite colourless. Purification is effected by crystallisation from glacial acetic acid and drying for 45 minutes at 80° C., and the reagent is kept in a vacuum desiccator in the dark. The determination of creatinine in a 5 per cent. solution of broth-cubes (defatted) or a 0.5 per cent. solution of meat-extract is made as

follows:—Evaporate 50 ml., after addition of 20 ml. of *N* hydrochloric acid, in a porcelain dish on the water-bath. Dissolve the residue in water, neutralise, and make the volume up to 50 ml. Decolorise the solution in a cylinder by means of aluminium hydroxide. Add to 20 ml. of the filtrate 5 ml. of *N* hydrochloric acid and make up to 50 ml. Add to 2 ml. of this solution 4 ml. of a 2 per cent. solution of dinitrobenzoic acid in absolute alcohol and 1 ml. of 5 per cent. sodium hydroxide solution. At the same time prepare a standard solution (0.01 per cent. of creatinine in *N*/10 hydrochloric acid) and treat it similarly. Measure the colour in a Duboscq colorimeter or with a photo-electric colorimeter, preferably the latter. Take the readings of the instruments for different dilutions of the standard creatinine solution, and obtain the corresponding percentages of creatinine from a table. In broth cubes 0.32 to 0.8 per cent. of creatinine was found, and in meat extracts 5.3 to 6.3 per cent. The amounts in urine ranged from 0.082 to 0.193 per cent.

D. A.

Oil from the Seeds of *Canarium commune* L. A. Steger and J. van Loon. (*Rec. Trav. Chim. Pays-Bas*, 1940, **59**, 168–172.)—*Canarium commune* (*Burseraceae*) is a tree indigenous to the Moluccas, Java, the Celebes and Indo-China. The three-cornered oval fruits have three compartments, each containing an oily kernel resembling the sweet almond in form and taste and sometimes known as Java almonds. Samples of the oil were obtained by cold pressing in a small laboratory press and by extraction of the kernels with petroleum spirit. The kernels yielded 68.6 per cent. of oil (10.4 per cent. on the whole fruit). The press-cake contained 16 per cent. of water, 11.3 per cent. of mineral matter and 39 per cent. of crude protein; it is therefore a valuable fertiliser and cattle-food. The expressed oil had the following constants, and the corresponding figures for the extracted oil are given in brackets:—acid value, 0.22 (1.12); saponification value, 195.3 (194.4); iodine value (Wijs, 2 hours), 74.0 (68.8); iodine value (Wijs, 24 hours), 73.5 (70.2); Reichert–Meissl value, 0.64 (0.58); Polenske value, 0.43 (0.60); n_D^{70} , 1.4497 (1.4495); sp.gr. at 78°/4° C., 0.8740 (0.8728). The oil is solid at room temperature. It yielded 95.0 (93.8) per cent. of fatty acids soluble in petroleum spirit, 0.61 (0.24) per cent. of unsaponifiable matter, 4.4 (4.4) per cent. of glycerol residue C_3H_8 and 0.0 (1.54) per cent. of volatile and insoluble matter. The constants of the fatty acids were as follows:—iodine value (Wijs 2 hours), 76.6 (72.4); iodine value (Wijs, 24 hours), 77.1 (—); neutralisation value, 206.0 (205.1); mean molecular equiv., 272.3 (273.5); n_D^{70} , 1.4401 (1.4394); saturated acids per cent., (Bertram), 38.7 (40.7); mean molecular equiv. of the saturated acids, 263.3 (263.3). The fatty acids were separated into solid and liquid fractions by Twitchell's method, converted into ethyl esters, and fractionated by vacuum distillation. The results indicated the following composition (per cent.):—stearic acid, 9.7 (10.2); palmitic acid, 29.0 (30.5); 9-oleic acid, 38.3 (39.9); 9, 12-linolic acid, 21.8 (18.7); 9, 12, 15-linolenic acid, 1.2 (0.7). The statement of Grün and Halden (*Analyse der Fette und Wächse*, Vol. I, p. 378; Vol. II, p. 261) that canari oil polymerises when heated to 240° to 250° C. could not be confirmed, and it is suggested that these authors may have confused canari oil with Java-olive oil (Weder Meyer, *Z. Unters. Nahr. Genussm.*, 1906, **12**, 210; Abst., *ANALYST*, 1906, **31**, 361).

A. O. J.

Liver Oils of Some Terrestrial Animals. M. Tsujimoto and H. Koyanagi. (*J. Soc. Chem. Ind. Japan*, 1939, 42, 421–422B.)—The liver oils of (a) elephant, (b) dog, (c) rabbit and (d) hen, were obtained by extraction with alcohol, followed by ether, and subsequent treatment of the combined extracts with acetone. The oils had the following characteristics:

| | (a) | (b) | (c) | (d) |
|--|--------|--------|--------|--------|
| Sp.gr. at 30°/4° C. | 0.9137 | 0.9246 | 0.9222 | 0.9239 |
| n_D^{20} | 1.4695 | 1.4730 | 1.4660 | 1.4700 |
| Saponification value | 147.9 | 152.7 | 169.8 | 167.8 |
| Iodine value (Wijs) | 146.0 | 144.9 | 109.3 | 95.9 |
| Unsaponifiable matter, per cent. | 21.13 | 22.35 | 14.81 | 14.60 |
| Acid value | 7.0 | 4.5 | 8.1 | 2.3 |

Oils (b) and (d) contained appreciable amounts of vitamin A. Notable amounts of highly unsaturated acids were present in each oil. Palmitic, stearic and oleic acids were identified in the free acids from hen-liver oil. The unsaponifiable matter from each oil contained hydrocarbons. Hydrogen chloride addition products were formed by saturating the ethereal solutions with hydrogen chloride below 0° C. (a) The hydrochloride sintered above 120° C. and melted at about 130° C. with decomposition, and contained 29.7 per cent. of chlorine. (b) From 2.6 g. of unsaponifiable matter 0.7 g. of crude hydrocarbon was obtained (of iodine value 292.3) the highest yield yet observed; m.pt. over 115° C., with decomposition at 128–129° C., and chlorine content 30.15 per cent. (c) There was only a trace of precipitate of hydrochloride owing to the small amount of hydrocarbon present. (d) The m.p. of the hydrochloride was about 130° C. and its chlorine-content 29.83 per cent. All these hydrochlorides are similar to the analogous compounds obtained from pig, horse, sheep, ox and human liver, as also to the compounds from ishinagi, sperm and finback whales and dolphin liver oils. D. G. H.

Analysis of Chaulmoogra Oils. *Hydnocarpus anthelmintica* Oil and *Taraktogenos Kurzii* (Chaulmoogra) Oil. H. I. Cole and H. T. Cardoso. (*J. Amer. Chem. Soc.*, 1939, 61, 3442–3445.)—The oil of *Hydnocarpus anthelmintica* ranks next to that of *Hydnocarpus Wightiana* for the treatment of leprosy. The analysis (*J. Amer. Chem. Soc.*, 1938, 60, 614) was made by saponifying and liberating the fatty acids in the usual way, and separating the solid from the liquid acids by crystallisation from 80 per cent. ethyl alcohol. The two fractions, as ethyl esters, were fractionally distilled, and two final crystallisations for the separation of the liquid acids were made with 80 per cent. acetone to prevent formation of ethyl esters. The native name "Chaulmoogra" of *Taraktogenos Kurzii* oil has latterly been used to indicate any oil that contains chaulmoogric acid and so might be used for the treatment of leprosy. The collection of the seeds is difficult and dangerous, and usually a long time elapses before they can be expressed, so that the commercial oil is of poor quality, and is often adulterated, and has therefore fallen into some disrepute. Plantations would eliminate the difficulty, and in fact *Taraktogenos Kurzii* has been acclimatised in Brazil, whence came the oil used in this investigation. The high iodine values of certain fractions, which led previous investigators to suspect the presence of linolic or linolenic acids, has

now been found to be due to gorlic acid, which has two double bonds. If it is found that the value of the oil in the treatment of leprosy is due to the gorlic acid rather than to the other optically active fatty acids, then this, the true chaulmoogra oil, would be preferable to the other oils analysed. The keeping properties of the oil appear to be good, but, as with all the chaulmoogra oils, that from old seeds is liable to cause irritation.

| | Oils of: | | | | |
|--|------------------------------|----------------------------------|----------------------------|---------------------------------|------------------------|
| | <i>Hydnocarpus Wightiana</i> | <i>Hydnocarpus anthelmintica</i> | <i>Taraktogenos Kurzii</i> | <i>Carpotroche brasiliensis</i> | <i>Oncoba echinata</i> |
| Sp.gr. at 25/25° C. | 0.955 | 0.952 | 0.952 | 0.955 | — |
| Free fatty acid (as oleic), per cent. | 2.7 | 2.9 | 1.3 | 3.6 | 4.3 |
| Saponification value | 201.0 | 203.3 | 200.6 | 201.8 | 193.7 |
| Iodine value (Hanus) | 98.4 | 89.2 | 101.5 | 108.0 | 96.4 |
| Spec. optical rotation $[\alpha]_D^{25}$ | 55.0 | 49.7 | 49.8 | 53.8 | 51.7 |
| Refractive index, n_D^{25} | 1.4799 | 1.4772 | 1.4790 | 1.4790 | — |
| Unsaponifiable matter, per cent. | 0.25 | 0.50 | 0.29 | — | — |
| Percentage composition of acids— | | | | | |
| Hydnocarpic | 48.7 | 67.8 | 34.9 | 45.0 | none |
| Chaulmoogric | 27.0 | 8.7 | 22.5 | 24.4 | 74.9 |
| Gorlic | 12.2 | 1.4 | 22.6 | 15.4 | 14.7 |
| Lower homologues of | | | | | |
| hydnocarpic | 3.4 | 0.1 | 0.4 | ? | ? |
| Oleic | 6.5 | 12.3 | 14.6 | 6.3 | 2.2 |
| Palmitic | 1.8 | 7.5 | 4.0 | 6.6 | 7.8 |
| Loss | 0.4 | 2.2 | 1.0 | 2.3 | 0.4 |

D. G. H.

Quantitative Estimation of Coloured Components in Paprika. L. Cholnoky. (*Z. Unters. Lebensm.*, 1939, 78, 157–161.)—For the commercial evaluation of paprika a knowledge of the proportions of the different paprika pigments is important. Red, ripe paprika contains the following polyene colour components:—capsorubin ($C_{40}H_{60}O_4$), capsanthin ($C_{40}H_{58}O_3$), zeaxanthin ($C_{40}H_{56}O_2$), cryptoxanthine ($C_{40}H_{56}O$), β -carotene ($C_{40}H_{56}$) and a trace of α -carotene ($C_{40}H_{56}$). The polyene alcohols are present as esters of higher fatty acids. Capsicum red is a coloured wax. Capsanthin and capsorubin esters, especially the former, are responsible for the bright red colour of paprika. The percentages of the different colour components may vary widely, and hence an estimation of the total colour is not sufficient for the evaluation. The only accurate method is to estimate the separate colour components by applying the chromatographic adsorption process of Tswett (*cf.* L. Zechmeister and L. Cholnoky, *Die chromatographische Adsorptionsmethode*, Vienna, 1938). For the extraction, 0.5 to 1.5 g. of the ground sample are transferred to a funnel, provided with a cottonwool plug of 3 to 5 mm. The colour is extracted with boiling petroleum spirit (b.p. 60 to 70° C.) in successive portions of 10 ml., and the extraction is continued until the solvent remains colourless. About 100 to 150 ml. of petroleum spirit are required. The separation is made in an adsorption tube (23×3.5 cm.), the lower part of which is filled with calcium hydroxide and the upper half with calcium carbonate. The latter adsorbs

the capsorubin and capsanthin esters, the former the yellow components. The chromatogram shows layers due to the separate colours, which may be measured in a colorimeter. The average amounts of coloured components per kg. found in 19 samples of paprika from different crops were as follows: carotene, 0.52; cryptoxanthin, 0.16; zeaxanthin, 0.41; capsanthin, 2.19; capsorubin, 0.49; total, 3.77 g. The paprika fruit-shells are usually ground up with 40 to 70 per cent. of seeds. The above-mentioned components estimated in 18 samples as supplied by the factories, amounted on the average to 0.31, 0.10, 0.20, 1.28 and 0.27 g., respectively; total 2.16 g. per kg. The loss of colour on keeping was also determined; for pure paprika it amounted to 0.86 per cent. after 6 months and 8.56 per cent. after 12 months. For 100 kg. of paprika ground with 40 kg. of seeds the losses after 6 and 12 months were 5.45 and 12.73 per cent. of the initial colour, and when the same mixture was stirred daily, the losses after 1, 2 and 3 months were 24.54, 35.34 and 48.48 per cent., respectively. Under normal conditions the pigment-content is not affected much. Pure polyenes are very sensitive to oxygen, and as a rule are destroyed within 1 to 2 months, with decolorisation. That this is not so with paprika pigments may be explained by their enclosure in lipoids, which protect them from oxidation. Thus paprika is a good stable source of provitamin A.

D. A.

Tests for Tannic Acid. D. B. Dott. (*Pharm. J.*, 1940, 144, 137.)—The solubility of 0.5 g. of tannic acid in 10 ml. of acetone is recommended as a test for the purity of the sample, and the colour and turbidity of a solution of 0.5 g. of the acid in 10 ml. of collodion (meth.) as a test for the suitability of the specimen for hospital use. The following procedure is suggested for quantitative examination:—Cupric sulphate (1.5 g.) is dissolved in 40 ml. of water, and the solution is added to a solution of 1 g. of the tannic acid in 20 ml. of water. A solution of crystalline sodium acetate (1.7 g.) in 10 ml. of water is added, and the liquid is finally treated with 0.5 ml. of acetic acid and mixed. After 3 or 4 hours the precipitate is collected on a weighed filter, washed free from sulphate and dried at about 100° C. to constant weight. These tests were applied to 4 samples of tannic acid which had passed the B.P. tests but had proved to be unsatisfactory in hospital practice, with the following results:

| Sample | Collodion solution | Solubility in acetone | Weight of copper ppt. g. |
|--------|--|--|--------------------------|
| A | Very turbid | Considerable amount insoluble | 1.143 |
| B | Almost clear, slowly giving slight deposit | Nearly clear, slowly giving small quantity of brown sediment | 1.179 |
| C | Clear straw-colour, no deposit | Clear, almost colourless, faint straw colour but no deposit after 24 hours | 1.163 |
| D | Clear brownish colour, soon changing to objectionable green colour | — | — |

E. M. P.

New Solanaceous Alkaloids from *Duboisia myoporoides*. W. Mitchell. (*Pharm. J.*, 1940, 144, 137.)—"Duboisine," the total alkaloids extracted from *Duboisia myoporoides*, has previously been found to contain hyoscyne and four new

alkaloids, tigloidine, valeroidine, poroidine, and isoporoidine (*J. Chem. Soc.*, 1937, p. 1820; 1938, p. 1685); there is no trace of hyoscyamine or other similar alkaloid. The four new alkaloids have now been identified. Tigloidine is tiglyl- ψ -tropëine; it has been synthesised. Valeroidine is the mono*iso*valeryl ester of a dihydroxy-tropeine previously isolated from Peruvian coca leaves as the dibenzoyl ester, and a structural formula has been suggested. Poroidine and isoporoidine have been shown to be *iso*valerylnortropëine and *d*- α -methylbutylnortropëine, respectively, and both have been synthesised. They have been isolated in the form of a mixture, known as base Z, which has been partly separated by an indirect method. *Iso*-valerylnortropëine has been isolated from this mixture, and although *d*- α -methylbutylnortropëine has not been isolated, its presence is practically certain. A mixture of 10 parts of the former with 1 of the latter has been found to resemble base Z closely.

E. M. P.

Rapid Method for the Determination of Nicotine. A. Verda and E. Herzfeld. (*Z. anal. Chem.*, 1939, 118, 9-13.)—Existing methods, which are reviewed, are considered troublesome and lengthy. The following procedure, which depends on the nephelometric measurement of the turbidity produced when nicotine reacts with silicotungstic acid in presence of gum arabic, requires only 60 to 90 minutes. A mixture of the tobacco (5 to 20 g., according to the nicotine-content), 2 g. of magnesium oxide, 30 g. of sodium chloride and 300 ml. of water is steam-distilled, and 300 ml. of distillate are collected in a receiver containing 3 g. of gum arabic; this ultimately dissolves in the distillate, which is then filtered through glass wool. A standard solution is prepared by dissolving 1 g. of pure nicotine in 100 ml. of a 1 per cent. solution of gum arabic (which has been filtered through glass wool), and 10 ml. of this solution are diluted to 100 ml. with the same gum arabic solution. Volumes of this solution corresponding with 5, 2.5, 1.25, etc., down to 0.0391 mg. of nicotine are then pipetted into each of 8 nephelometer tubes, and the volume is made up to 5 ml. (where necessary) with the gum arabic solution. The reagent is prepared by dissolving 1 g. of gum arabic and 10 g. of silicotungstic acid in a mixture of 50 ml. of water and 50 ml. of glycerol; if the solution is allowed to stand overnight and is then filtered through glass wool, it keeps well. On the addition of 0.5 ml. of reagent to the solutions containing the nicotine, a milky turbidity is produced which remains unchanged after 1 day and cannot be separated in the centrifuge, and this is used to match the sample against the series of standard tubes, the sensitiveness of the method being 0.31 mg. of nicotine; it may therefore be used to recognise "nicotine-free" tobaccos having a limiting nicotine-content of 0.1 per cent. Up to 10 per cent. of pyridine or ammonia does not interfere, and the accuracy of matching is unaffected by the yellow colour of the mixture observed in transmitted light. The recorded errors for experiments in which 15 to 100 mg. of nicotine were used varied from +5 to -7 mg.; in general, the higher the quantity of nicotine present, the greater the negative error.

J. G.

Determination of Nicotinic Acid Diethylamide and Phthalic Acid Bis-diethylamide. K. A. Jackerott and F. Reimers. (*Z. anal. Chem.*, 1939, 117, 415-420.)—The method is based upon the hydrolysis of the acid amide into its constituent acid and diethylamine, and determination of the diethylamine by

distillation into a standard acid solution. Preliminary experiments showed that nicotinic acid diethylamide (coramine) is completely hydrolysed when boiled for 30 minutes with 5 *N* sodium hydroxide solution. The method is as follows:—Nicotinic acid diethylamide (0.4 g. or a corresponding amount of its preparations) is mixed in a distillation flask with 100 ml. of water and 100 ml. of conc. sodium hydroxide solution, and after the addition of pumice the flask is connected with a Kjeldahl distillation apparatus. The receiver contains 25 ml. of 0.1 *N* hydrochloric acid, and the orifice of the condenser should dip well beneath the surface of the liquid. The flask is heated at such a rate that its contents are maintained at boiling-point for 20 minutes with scarcely any distillation. The rate of heating is then increased, and 100 ml. of liquid are distilled. Then 25 ml. of water are introduced by means of a separating funnel fitted into the stopper of the flask, and distillation is continued until the distillate measures 125 ml. The liquid in the receiver is titrated with 0.1 *N* sodium borate or sodium hydroxide solution, methyl red being used as indicator, and the result of a blank determination (obtained by distillation of a mixture of 100 ml. of water and 100 ml. of sodium hydroxide solution) is deducted. Each ml. of 0.1 *N* hydrochloric acid is equivalent to 0.01781 g. of nicotinic acid diethylamide. In recent years commercial preparations with constituents resembling nicotinic acid diethylamide in constitution and pharmacological properties have been sold. The application of the method to one of these compounds, *viz.* phthalic acid bis-diethylamide (Neospiran) was investigated. Preliminary experiments showed that hydrolysis of this compound is effected more rapidly by dilute hydrochloric acid than by alkali. The method recommended is as follows:—The sample (0.3 g.) is heated under a reflux condenser with 20 ml. of dilute hydrochloric acid for 30 minutes. When cold the product is transferred to the distillation flask with 80 ml. of water, and, after the addition of 100 ml. of conc. sodium hydroxide solution, the liberated diethylamine is distilled into 0.1 *N* hydrochloric acid in the manner previously described. The blank determination is made upon a mixture of 20 ml. of dilute hydrochloric acid, 80 ml. of water and 100 ml. of the sodium hydroxide solution. Each ml. of 0.1 *N* hydrochloric acid is equivalent to 0.01381 g. of phthalic acid bis-diethylamide. Unlike nicotinic acid diethylamide, phthalic acid bis-diethylamide is hydrolysed only slowly in alkaline solution. It is therefore necessary to ensure complete hydrolysis before the liquid is transferred to the distillation flask. The time of 30 minutes for hydrolysis may be exceeded if required.

A. O. J.

Biochemical

Biochemical Behaviour of Lead. I. Influence of Calcium, Phosphorus and Vitamin D on Lead in Blood and Bone. A. E. Sobel, H. Yuska, D. D. Peters and B. Kramer. (*J. Biol. Chem.*, 1940, 132, 239–265.)—Experiments were carried out to determine the effect of calcium, phosphorus and vitamin D on the amount of lead present in the blood of young rats and on the deposition of lead in the bones. Groups of animals were fed on a low calcium—low phosphorus diet, a low calcium—high phosphorus diet, and a high calcium—low phosphorus diet all containing the same amount of lead, while some of the rats on each of the diets

were given vitamin D in addition. A satisfactory explanation of the results observed is that lead deposition is directed by a system of its own, this being governed by the same laws as calcium deposition, but not necessarily in the same direction. Thus there is an ideal lead—phosphorus ratio in the diet at each lead level that is most favourable for lead deposition in the bones, and the addition or removal of phosphorus decreases the amount of lead deposited. The addition of any agent, such as calcium, which behaves as if it removed phosphorus has the same effect. Vitamin D increases the deposition of lead with all types of diet by compensating for a disproportionate dietary ratio. The low calcium—low phosphorus diet was found to bring about the greatest degree of lead deposition. The effect of the three dietary factors on blood lead values may be similarly explained by assuming a relationship between blood-lead and dietary lead—phosphorus ratios analogous to the relationship between blood-calcium and dietary calcium—phosphorus ratios. As the lead—phosphorus ratio is increased the blood lead value is increased, so that the addition of phosphorus depresses the blood lead, whilst the addition of calcium, which has the effect of removing phosphorus, increases it. The effect of vitamin D on blood lead is similar to its effect on blood calcium, that is, blood lead is increased on a low calcium—low phosphorus diet, but is not markedly affected on a high calcium—low phosphorus diet or on a low calcium—high phosphorus diet.

F. A. R.

Molecular Weight of Egg Albumin. F. W. Bernhart. (*J. Biol. Chem.*, 1940, **132**, 189–193.)—Analyses were made of the phenylalanine, tyrosine and tryptophane contents of recrystallised, electro-dialysed egg albumin. From the results obtained the minimal molecular weight was estimated to be 18,400. When multiplied by 2 this gives a value of 36,800, which is in close agreement with the molecular weight found either by physical methods or by estimating the polysaccharide-content and assuming that one molecule is present in each molecule of the protein. On the assumption that no unknown basic amino acids or nitrogen-containing prosthetic group is present, the number of amino-acid groups in egg albumin was calculated to be 310 per molecule. This value approximates closely to the value of 288 calculated by Bergmann and Niemann.

F. A. R.

Estimation of Arginine by means of Flavianic Acid. H. B. Vickery. (*J. Biol. Chem.*, 1940, **132**, 325–342.)—A sample of the protein, weighing 25 g., or more according to the anticipated arginine-content, is hydrolysed by boiling for 24 hours with 500 ml. of 20 per cent. hydrochloric acid. The hydrolysate is repeatedly evaporated to a syrup under reduced pressure and then made up to 250 ml.; 1-ml. aliquot portions are removed for nitrogen determination, from which the amount of protein present is calculated. The solution is diluted, boiled with 5 g. of Norit and filtered, and the Norit is extracted twice with boiling water. The combined filtrate and washings are concentrated to 250 ml., and 50-ml. aliquot portions are taken for each of 4 arginine estimations. For single estimations the amounts of materials taken can be correspondingly reduced. For each mole of arginine present, calculated from the best estimate available, 4 to 5 moles of flavianic acid are added to the solution (1 g. of arginine requires 1.805 g. of flavianic acid for 1 mole) at room temperature. The samples are placed in the refrigerator

for 4 days, being thoroughly stirred at least once a day. Pale yellow needles of arginine diflavianate separate, occasionally in admixture with orange-yellow monoflavianate. The precipitate from each sample is filtered off on a small sintered funnel and washed with water saturated at room temperature with arginine monoflavianate. During this operation much of the diflavianate is converted into the monoflavianate. The precipitate is then stirred with a little hot water, 5 *N* ammonium hydroxide is added drop by drop from a 1-ml. pipette, and the precipitate is stirred until all is in solution; the minimal amount of ammonia is used. The solution is drawn through the filter into a filter-flask, and the filter is washed with hot water and, if necessary, a drop of ammonia. The filtrate is transferred to a beaker, the total volume being about 40 ml., and is then heated to boiling, and a slight excess of *N* sulphuric acid is added. Arginine monoflavianate usually crystallises at once from the hot solution, and crystallisation is allowed to become complete at room temperature before the solution is stirred. It is allowed to stand overnight in the refrigerator and then filtered on to a sintered glass crucible, and the precipitate is washed several times with water saturated at room temperature with arginine monoflavianate, and finally with a little alcohol. It is dried for a few hours at 105° C., cooled in a desiccator and weighed with as little exposure to the air as possible (it is hygroscopic). The weight of arginine monoflavianate, multiplied by 0.3566, gives the weight of arginine. Very good agreement was obtained in estimations carried out in quadruplicate on pure proteins, and in only one instance was the average variation greater than ± 1 per cent. The following are the results obtained with various proteins (per cent.): edestin, 16.76; tobacco-seed globulin, 16.09; cottonseed globulin, 14.92; arachin, 13.94; amandin, 13.31; gliadin, 2.57; zein, 1.60; casein, 3.72; haemoglobin (horse), 3.59; γ -pseudoglobulin (horse), 2.66; fibrin (cattle), 7.70; gelatin, 8.68; egg albumin, 5.66; hair (human), 9.37; wool (sheep), 10.4; silk fibroin, 0.76.

F. A. R.

Naphthalene- β -Sulphonic Acid as a Reagent for Amino Acids. M. Bergmann and W. H. Stein. (*J. Biol. Chem.*, 1939, **129**, 609–618.)—Naphthalene- β -sulphonic acid forms sparingly soluble salts with leucine, phenylalanine, arginine, histidine, tryptophane and cysteine. These salts, for which the term nasylates (not to be confused with the naphthalenesulphonyl or nasyl derivatives) is proposed, promise to be of great value in the isolation, purification and determination of these amino-acids. The determination of *l*-phenylalanine is given as an example. Three 15-ml. samples of a solution containing 0.906 g. of *l*-phenylalanine in 0.5 *N* hydrochloric acid were added to 1.004 g., 1.254 g. and 1.509 g. samples of ammonium nasylate (the most stable form of the reagent), each dissolved in 60 ml. of 0.5 *N* hydrochloric acid. The mixtures were continuously agitated at exactly 0° C. for 3 days, and the precipitates were collected at 0° C. on sintered glass filters, which were weighed, then dried to constant weight in a desiccator and re-weighed. The loss in weight enabled corrections to be applied for the weight of solid in the mother liquors retained by the precipitates. The amount of *l*-phenylalanine in the original solution was calculated from the corrected dry weights of the precipitates, and the average was found to be 99.3 per cent. of the quantity actually

taken. *l*-Arginine and *l*-histidine form sparingly soluble dinasylates as well as mononasylates. Some polypeptides also form sparingly soluble nasylates. All these salts appear to be well-crystallised compounds with definite, characteristic melting-points as follows: *l*-leucine mononasylate 187.5 to 189° C., *l*-phenylalanine mononasylate 232 to 233° C., *l*-arginine dinasylate 209 to 211° C., *l*-arginine mononasylate 243° C., *l*-histidine dinasylate 265° C., *l*-histidine mononasylate 206 to 207° C., glycyl-*l*-leucine mononasylate 211 to 212° C., all with decomposition. The amino acids can be regenerated by dissolving the salts in absolute alcohol, adding pyridine, allowing the mixture to stand for 2 days at room temperature, and filtering off the precipitated amino acid. This is purified by a second treatment.

F. A. R.

Experiments on Amino-Acids. I. Partition of Acetamino-acids between Immiscible Solvents. II. Separation of Amino-acids by means of their N-Acetyl Derivatives. III. Isolation of Hydroxy-amino-acids from Protein Hydrolysates. IV. Methyl Ethers of some N-Acetylhydroxy-amino-acids. R. L. M. Syngé. (*Biochem. J.*, 1939, 33, 1913–1917, 1918–1923, 1924–1930, 1931–1934.)—I. The partition coefficients of a number of acetamino-acids between chloroform and water, ether and water, and ethyl acetate and water, were measured, and very wide variations were found, suggesting a possible basis for the separation of amino-acids. II. A mixture of 14 amino-acids, intended to simulate a gelatin hydrolysate, was acetylated at 0° C. with acetic anhydride and sodium hydroxide solution. The solution of acetylated acids was made slightly acid with sulphuric acid and extracted with chloroform in a Neuberger continuous extractor. The aqueous phase, which contained about 20 per cent. of unchanged amino-acids, was re-acetylated. After a second re-acetylation, the three chloroform extracts were examined for individual amino-acids. Amino-acids susceptible of separation by extractional fractionation were obtained in high yield, namely, leucine, phenylalanine, proline, methionine, valine, alanine and glycine. III. Serine and hydroxy-proline were acetylated separately by means of acetic anhydride and alkali, as already described. The resulting alkaline solutions were allowed to stand for some time to ensure hydrolysis of any O-acetyl groups, and were then neutralised and treated with benzoyl chloride at 0° C. in the absence of excess alkali. Good yields of N-acetyl-O-benzoyl-amino acids were obtained, and these were readily extractable from aqueous solution into chloroform; on treatment with dilute aqueous alkali at room temperature they were rapidly converted into N-acetyl derivatives. A protein hydrolysate, after being freed from bases by precipitation with phosphotungstic acid, was acetylated and, after being kept alkaline overnight, was benzoylated as described above. The mixture, containing acetamino-acids, N-acetyl-O-benzoyl-hydroxy-amino-acids and benzamino-acids, was extracted with chloroform in a Neuberger continuous extractor. The chloroform was removed from the extract, and the residue dissolved in dilute alkali solution at room temperature. This saponified the O-benzoyl group only, and on acidifying the solution and exhaustively extracting with chloroform, a solution containing N-acetyl-hydroxy-amino-acids was obtained. The free amino-acids were prepared by acid hydrolysis. A preliminary fractionation of hydrolysates of

fibrin, wool and gelatin was attempted by the method. IV. The individual components of the hydroxy-amino-acid fraction were not readily separable from one another by direct crystallisation. Accordingly, a number of N-acetyl-hydroxy-amino-acids were methylated by means of silver oxide and methyl iodide, and the methyl esters of the N-acetyl-O-methyl-hydroxy-amino acids were isolated by distillation and hydrolysed to the free acid. The partition coefficients between chloroform and water of the compounds derived from *dl*-serine, *dl*-allothreonine *l*-hydroxyproline and *l*-tyrosine were sufficiently different to suggest that this procedure provided a basis for fractionating a mixture of these amino-acids, but the separation has not yet been attempted.

F. A. R.

Colorimetric Estimation of Quinine in Biological Fluids and Tissues.

R. O. Prudhomme. (*J. Pharm. Chim.*, 1940, 1, 8-17.)—When a few drops of a 2 per cent. solution of eosin are added to a 1 per cent. solution of a quinine salt a red precipitate is formed which, when shaken with chloroform, dissolves to form an intensely red solution. The colour is perceptible when the amount of quinine present is only 1/1000 mg. in 10 ml. of water. The reaction is given by other alkaloids, *e.g.* cinchonine, ephedrine, eserine, pilocarpine and atropine, but not by caffeine. The colour attains its maximum intensity at pH 6.5 to 7.5, and this condition may be secured by the use of a buffer solution made by mixing 13.617 g. of monopotassium phosphate dissolved in 300 ml. of water with 41.566 g. of disodium phosphate dissolved in 700 ml. of water. Standards for the colorimetric estimation of quinine in urine are prepared by adding amounts of quinine sulphate ranging from 0.01 to 2 mg. to 10-ml. portions of normal urine. Each portion is treated with 5 ml. of a 10 per cent. lead acetate solution and filtered. The filtrate (12 ml.) is treated with 5 drops of conc. sulphuric acid and filtered repeatedly until clear, and 9 ml. are neutralised to the lilac end-point of litmus solution with sodium hydroxide solution. The mixture is treated with 2 ml. of the phosphate buffer solution and 4 drops of a 2 per cent. eosin solution and is finally shaken thoroughly with 3 ml. of chloroform. After 4 hours the chloroform layer is transferred by means of a pipette to a tightly stoppered tube. Sealed tubes may be used, but care should be taken that the portion of the tube to be heated is not contaminated with chloroform, the decomposition products of which affect the colour of the solution. The urine to be investigated is treated in the same manner. Comparison of sample with standard should not be made until the chloroform layer has been separated and transferred to a clean tube since the inclusion of the supernatant liquid between the chloroform layer and the walls of the tube may lead to erroneous results. For the determination of quinine in blood and serum, the standards are prepared as follows:—The coagulating power of the blood is destroyed by addition of powdered potassium oxalate, and amounts of quinine sulphate ranging from 0.005 to 0.1 mg. are added to 10-ml. portions. Each portion is treated with 20 g. of crystalline sodium sulphate and 7 ml. of *N* sulphuric acid, and the mixture is heated in a water-bath at 45° to 50° C. for 30 minutes with occasional shaking. The brown solution is filtered, and the filtrate is cooled until the sodium sulphate crystallises. The supernatant liquid (10 ml.) is neutralised with sodium hydroxide solution, phenolsulphonephthalein being used as external

indicator, and is treated with 2 ml. of the buffer solution, 6 drops of eosin solution and 3 ml. of chloroform. After being vigorously shaken the mixture is allowed to stand for 6 hours, and the chloroform layer is transferred to a stoppered tube. The sample of blood to be tested is treated with potassium oxalate and subjected to the same process. For organs or tissue the procedure is as follows:—The material (10 g.) is finely ground in a mortar with 30 g. of crystalline sodium sulphate and, if necessary, a little sand. The paste so obtained is rinsed into a flask with 10 ml. of water, 10 ml. of *N* sulphuric acid are added, and the mixture is heated for 30 minutes at 45° to 50° C. The method is then as described for blood. To obtain standards different amounts of quinine sulphate are added to similar material in the mortar. It is important to notice that with normal fresh organs a blank estimation may give results corresponding with 0.01 mg. of quinine. It is not possible therefore to determine less than this amount, and when putrefaction has set in the colour given in a blank estimation may be intense. Although not specific, the reaction is useful for the study of the distribution of quinine and other alkaloids in the organs and their rate of excretion. The reaction is not given by aspirin, the sulphonamides, veronal and gardenal, nor by excretory substances such as urea and the amino acids. The red compound was isolated and was found to have no bitter taste and to be devoid of the therapeutic properties of quinine for malaria. A study of several of the eosin-alkaloid compounds showed that they can be distinguished by their ultra-violet absorption spectra—a method that may prove useful in toxicological practice.

A. O. J.

Photometric Estimation of Silicic Acid in Biological Substances.

J. Bodnár and T. Török. (*Z. physiol. Chem.*, 1939, 261, 257–268.)—The chief difficulty in the colorimetric estimation of silica as silicomolybdic acid is the interference caused by the presence of iron and phosphate. In the method described this is overcome by converting the iron into a water-soluble complex and precipitating the phosphate with magnesia mixture. From 0.05 to 0.25 g. of the powdered tissue is weighed into a small platinum crucible and mixed with 0.25 g. of fusion mixture (potassium and sodium carbonates). The crucible is heated over a Teclu burner, gently at first, but with gradual increase of the temperature as the organic matter becomes charred, and is finally ignited for 40 minutes (not less) with the full heat of the burner. The fused mass is stirred at intervals with a platinum wire. After cooling, the melt is dissolved in water, 1 to 2 drops of methyl orange solution are added, and the solution is made slightly acid by neutralising with 3 per cent. hydrochloric acid and adding 0.5 ml. in excess. The solution is transferred to a 15-ml. graduated flask and treated in turn with 0.5 ml. of 6.5 per cent. potassium cyanide solution and 1.5 ml. of acid magnesia mixture (42 g. of ammonium chloride, 22 g. of magnesium chloride, 1 g. of citric acid and 1.3 g. of ammonium dihydrogen phosphate in 200 ml. of 3 per cent. hydrochloric acid). Three ml. of 8 per cent. ammonia solution (prepared from ammonium chloride, the aqueous vapours being condensed and collected in an apparatus protected by a film of paraffin) are added dropwise to the solution, which is constantly shaken. The solution is then diluted to the mark, transferred to a centrifuge tube, and centrifuged for 5 to 10 minutes. Ten ml. of the supernatant liquid are transferred

to a 25-ml. graduated flask with the aid of a pipette and treated with 1.5 ml. of ammonium molybdate solution (10 g. of the salt are dissolved in 200 ml. of 5 per cent. sulphuric acid, and the solution is filtered after standing for 24 hours) and 1.5 ml. of hydroquinone solution (4 g. of hydroquinone in 200 ml. of water containing 4 drops of conc. sulphuric acid) and thoroughly shaken. After standing for 5 minutes the blue solution is diluted to the mark with sodium sulphite solution (made by dissolving 30 g. of anhydrous sodium sulphite in 200 ml. of water and 160 g. of anhydrous sodium carbonate in 800 ml. of water, and mixing the two filtered solutions) and again shaken. The solution is transferred to the cell of the photometer and exposed to the light of the photometer for 10 minutes, and the reading is taken, a red filter being used. The extinction value of a solution prepared in the same way from the reagents only is measured and subtracted from that of the test solution. With pure silica solutions the error was never in excess of 2 per cent., whilst in presence of different amounts of iron and phosphorus the greatest error found was + 6.7 per cent. In estimations of the silica-content of organic materials, duplicate experiments agreed usually within ± 2 per cent., whilst the error in the recovery of added silica never exceeded ± 5 per cent.

F. A. R.

Fatty Acid Dehydrogenase and its Co-enzyme. O. St. A. K. Lang and H. Mayer. (*Z. physiol. Chem.*, 1939, 261, 249–252.)—Extracts of liver and muscle-tissue brought about dehydrogenation of saturated fatty acids when incubated with the potassium salts of those acids in presence of methylene blue and a co-enzyme preparation. The product obtained from palmitic acid was a hexadecenoic acid, presumed to be the $\alpha\beta$ -unsaturated acid, though the position of the double bond is not certain. The dehydrogenation of oleic acid proceeded more slowly, and of linolic acid more slowly still. The co-dehydrogenase was shown to be identical with muscle adenylic acid.

F. A. R.

On the Fatty Acid Dehydrogenase IV. Product of the Dehydrogenation of Stearic Acid. O. St. A. K. Lang and F. Adickes. (*Z. physiol. Chem.*, 1939, 262, 123–127.)—In an earlier paper it was stated that stearic acid is dehydrogenated by the enzyme to the corresponding $\alpha\beta$ -unsaturated acid. This is not correct, the product being oleic acid, as was shown by the isolation (as the 2 : 4-dinitrophenylhydrazones) of azelaic aldehydic acid from the product of ozonolysis. F. A. R.

Estimation of Vitamin C by means of the Zeiss Step-Photometer. F. Bukatsch. (*Z. physiol. Chem.*, 1939, 262, 20–28.)—The usual method of estimating ascorbic acid by titration with 2 : 6-dichlorophenolindophenol in strongly acid solution suffers from three disadvantages. First, the end-point may not be very sharp, especially with biological fluids such as tissue-extracts and press-juices. Secondly, unless the titration is carried out very rapidly, large errors are introduced because of fading of the indicator. Thirdly, the method is not applicable to certain extracts containing plant-pigments. All these disadvantages are overcome in the method now proposed, and good results are recorded. About 3 g. of the fresh material, e.g. leaves, are minced and ground in a mortar with sand

and 3 ml. of 20 per cent. metaphosphoric acid. The resulting paste is diluted to 30 ml. with air-free water and filtered. An aliquot portion of the filtrate is transferred to a test-tube and 5 ml. of pure nitrobenzene are added. A quantity of 2 : 6-dichlorophenol indophenol solution equivalent to 0.5 mg. of ascorbic acid is added from a pipette, and the tube is gently inverted. The colour of the dye slowly fades and may disappear, in which event a further quantity of indicator solution should be added. When no further fading occurs the tube is vigorously shaken to transfer the remainder of the indicator to the nitrobenzene phase. Xylene may be used instead of nitrobenzene and the indicator must then be introduced below the xylene layer by means of a long pipette. The colour of the dye in either solvent remains practically unchanged for at least ten minutes, and the extinction value of the solution is measured in the 0.5-cm. cell of a Zeiss step-photometer, filter S53 being used with nitrobenzene and S50 with xylene. By using solutions of pure ascorbic acid the relationship between the extinction value of the indicator solutions and the concentration of ascorbic acid was shown to be linear in both instances. From the resulting graph it is possible to calculate the ascorbic acid content of the solution under examination. Even bright red extracts, such as are obtained from red cabbage leaves or beetroot, give satisfactory results by this method, for the red pigments are not extracted by either nitrobenzene or xylene.

F. A. R.

The Red Oxidation Products of the Tocopherols. W. John and W. Emte. (*Z. physiol. Chem.*, 1939, 261, 24-34.)—Furter and Meyer (*cf. ANALYST*, 1939, 64, 217) made use of the intense red colour produced by heating tocopherols with nitric acid in their method of assaying vitamin E preparations. It has now been found that α - (or β -) tocopheryl quinone, which is the first oxidation-product, is further oxidised to a red substance, which is itself converted into another compound by prolonged treatment. Thus, in Furter and Meyer's method, not only are the biologically active tocopherols converted into the red substance, but also the inactive tocopheryl quinones. This observation would account for the fact that Emmerie and Engel (*cf. ANALYST*, 1939, 64, 446) obtained higher values by the chemical method of assay than by the biological method.

F. A. R.

Fluorescence Analysis of Human Urine. W. Koschara, S. von der Seipen and P. A. Aldred. (*Z. physiol. Chem.*, 1939, 262, 158-167.)—Urine itself does not fluoresce in daylight, but if it is stirred with active charcoal, pigments are adsorbed from urine; by eluting the charcoal with hot 0.02 *N* sodium hydroxide solution, an eluate is obtained that exhibits a blue fluorescence. Uropterin, the chief fluorescent pigment of urine, is quantitatively recovered from a solution by such treatment, but the fluorescence of the eluate from urine is not due entirely to uropterin, solutions of which have a blue-green fluorescence. Uropterin is selectively adsorbed from 0.1 *N* hydrochloric acid solution on to bleaching-earth, from which it is eluted by aqueous pyridine, and by applying this technique to the charcoal eluates from a large number of specimens of urine it was shown that, in general, one-third to one-half of the fluorescence is due to uropterin. The charcoal adsorption and elution method was applied to a large number of

urines voided by patients suffering from many different diseases, and an attempt was made to assess the clinical value of the uropterin estimation. The results are regarded as confirmation of the authors' hypothesis, that the uropterin content of urine is a measure of oxidative nitrogen metabolism. F. A. R.

Bacteriological

Silicic Acid Nutrient Media. O. Hettche. (*Zent. f. Bakt.* I. Abt. Orig., 1939, 5, 144; *Bull. Hyg.*, 1940, 15, 48.)—The author has previously reported on the value of silica jelly for bacterial cultivation. He here describes a simplification of this medium, water-glass being used as a substitute for agar or gelatin. By using 0.05 per cent. sodium sulphite and Cenovis (a yeast and vegetable extract), peptone can be dispensed with, and it is claimed that the size of the colonies is half as large again as on ordinary nutrient agar. The latest development is the use of 0.5 per cent. of peptone and 0.1 to 0.2 per cent. of Cenovis with 7 per cent. of water glass. Growth on this medium compares favourably with that on lactose agar. When sodium sulphite is used and the pH is adjusted to 7.2 the medium sets in about 6 minutes at 20° C. D. R. W.

Toxicological

Extraction of Alkaloids with Acetone. P. Cheramy and M. Papavassilou. (*J. Pharm. Chim.*, 1940, 30, 316–321.)—A method based on that of Cheramy and Lobo for the extraction of barbiturates (*J. Pharm. Chim.*, 1934, 20, 400–403; Abst., *ANALYST*, 1935, 60, 50) is satisfactory for the extraction of alkaloids from viscera. The pulped organs are heated for two hours on the water-bath with 3 volumes of acetone in presence of tartaric acid, the solvent is removed (without heat), and the extraction is repeated. The residue is washed with acetone, and the combined acetone extracts are cooled to 0° C., filtered and distilled, the distillation being begun at normal pressure and finished under reduced pressure. The syrupy residue is dissolved in 300 ml. of warm anhydrous acetone, re-cooled, filtered through a filter moistened with acetone, and washed with the solvent. After distillation of the acetone, the soft residue is dissolved in 100 to 200 ml. of 20 per cent. ammonium sulphate solution, filtered cold and exhausted with various volatile solvents in acid and then in alkaline solution in the usual way. No appreciable loss of alkaloid occurs through the precipitation of impurities with ammonium sulphate solution. For some alkaloids (*e.g.* quinine and strychnine) which give too low results by this and by the Stas-Otto method, results of test determinations *in vitro* and *in vivo* have been greatly improved through the replacement of the first extraction on the water-bath by a Soxhlet extraction with acetone acidified with acetic acid; *e.g.* strychnine in egg, from 57 to 80 per cent. of theory, quinine in beef muscle, from 83 to 92 per cent. of theory. Other results of test analyses are given. E. B. D.

Gas Analysis

Separation of Para-Hydrogen from Oxygen and Carbon Monoxide. E. Bergmann, L. Farkas and L. Sandler. (*J. Amer. Chem. Soc.*, 1940, **62**, 445.)—Since *para*-hydrogen is often used as an aid to the elucidation of reaction mechanisms, it is necessary to determine the *para-ortho* ratio of the hydrogen, and therefore, to separate the hydrogen from all other gases concerned in the reaction in such a way that the *para-ortho* ratio remains unchanged. Most of the common gases can be separated by liquefaction with the aid of liquid air, but liquid hydrogen is necessary for carbon monoxide or oxygen, and, as it is not always available and is expensive to prepare, the following alternative methods are proposed:—Oxygen may be absorbed quantitatively in a saturated solution of triphenylmethyl sodium in dibutyl ether containing an excess of the solute; dibutyl ether is preferable to the diethyl ether recommended by Schlenk and Marcus (*Ber.*, 1914, **47**, 1664), as it has a lower vapour-pressure and will not dissolve the grease from stopcocks. The solution, prepared in the usual way (*loc. cit.*) and filtered, is then evaporated in a stream of nitrogen, dibutyl ether being added gradually during the process. The resulting solution is stable for many months if stored *in vacuo*, and if it is cooled its rate of absorption is very high. In one experiment a closed system of 5-ml. capacity was connected through a narrow capillary tube with a wash-bottle containing 20 ml. of the solution; when 130 cb. mm. of oxygen (at S.T.P.) had passed through the capillary tube, the residual oxygen was no longer measurable (*i.e.* it was less than 0.005 per cent.). The alteration in the *para-ortho* ratio of the hydrogen during the determinations was negligible, and was probably due to the presence of small quantities of free (paramagnetic) triphenylmethyl. The usual ammoniacal cuprous oxide solution is recommended for the absorption of carbon monoxide, and when under the conditions described above 500 cb. mm. (S.T.P.) of the gas (actual pressure, 70 mm.) had passed through the tube, less than 0.005 per cent. remained unabsorbed. Here, again, a negligible change in the *para-ortho* ratio was observed, and this is attributed mainly to divalent (*para*-magnetic) copper compounds. These methods may also be useful for other hydrogen modifications, *e.g.* mixtures of *ortho*-hydrogen and deuterium (*cf.* *J. Amer. Chem. Soc.*, 1939, **61**, 3393). J. G.

Agricultural

New Derivatives of Constituents of Derris Root. Th. M. Meyer and D. R. Koolhaas. (*Rev. Trav. Chim. Pays-Bas*, 1939, **58**, 1119–1123.)—(A) By the action of alcoholic potash on derride, 4-hydroxycumarone-5-carboxylic acid was formed (*cf.* Manjunath, Seetharamiah and Siddappa, *Ber.*, 1939, **72**, 93). (B) Oxidation of an alkaline solution of derridenone with hydrogen peroxide, by the Späth and Pesta method (*Ber.*, 1934, **67**, 853), yielded furan-2-3-dicarboxylic acid. (C) Rissic acid and (D) 2-hydroxy-4,5-dimethoxybenzoic acid were among the products of oxidation of dehydroderride with potassium permanganate in acetone. Full experimental details are given. E. B. D.

Water

Determination of Iodide in Mineral Waters containing Bromide and large quantities of Chloride. E. Müller and W. Stumpf. (*Z. anal. Chem.*, 1939, 118, 90–93.)—Determination of iodine in mineral waters containing much chloride (1:100,000) by the usual method (oxidation with sodium nitrite and hydrochloric acid) gives inconsistent results. After the oxidation the carbon disulphide is not always violet, but, according to the conditions, more or less brown, so that the results are too low. It was found that oxidation with very small quantities of nitrite and of hydrochloric acid yielded comparative figures. Although all the iodine is not extracted, a factor can be used for calculating the true iodine-content. It is essential to keep the carbon disulphide constantly covered with water during the washings. The procedure is as follows:—(1) Approximate determination of the iodine-content without using a factor. (2) Preparation of a type-solution, containing the same proportion of salts as the water to be analysed and with the same iodine-content as found; from the results obtained in the iodine determination the factor to be applied can be calculated. (3) Accurate determination of the iodine-content, applying the factor found. Iron, manganese or bromine do not interfere; hydrogen peroxide oxidises a small amount of bromide very slowly after long shaking.

D. A.

Organic

Preparation of Grignard Reagents from Magnesium Amalgams. E. G. Rochow. (*J. Amer. Chem. Soc.*, 1939, 61, 3591.)—The phase diagram for the system Mg-Hg shows two compounds, MgHg_2 and MgHg . Below 168°C . the equilibrium condition at the low magnesium end is a mixture of MgHg_2 crystals and liquid, so that the reported solubility of magnesium in mercury (3 per cent. at 250°C ., 1 per cent. at 100°C . and probably 0.1 per cent. at room temperature) must refer to the solubility of MgHg_2 . Amalgams of 5.71 per cent. or greater magnesium concentration will be solids; those with 5 to 0.1 per cent. of magnesium will normally be mixtures of MgHg_2 with increasing proportions of liquid. Amalgams containing from 0.1 to 1.0 per cent. of magnesium were prepared in an all-glass apparatus under an atmosphere of purified nitrogen, and after cooling in nitrogen, 50 ml. of 0.1 *N* solution of methyl magnesium chloride were added through the condenser to eliminate difficulties in starting, and methyl bromide was admitted. After refluxing for some hours samples were withdrawn for determination of total CH_3MgX by evolution of methane with water, and the increase of CH_3MgX over that added at first was calculated as percentage yield based on the magnesium. Typical examples were:—Percentage of magnesium in amalgams, 0.1, 0.5 and 1.0 gave respectively 0, 4.1 and 25.3 yield of RMgX per cent. The yield of RMgX from magnesium amalgams increases with concentration of magnesium in the amalgam with increasing possibility of free magnesium in the amalgam mixture, and it is concluded that MgHg_2 does not participate in the Grignard reaction as readily as magnesium, and that mercury has therefore an inhibiting effect.

D. G. H.

Gradual Decomposition by Oxidation of Fatty Acids into their next lower Homologues. H. Mendel and J. Coops. (*Rec. Trav. Chim. Pays-Bas.*, 1940, 58, 1133–1143.)—A new method for the gradual decomposition of fatty acids into their next lower homologues has been tested on palmitic and stearic acids. The steps of the decomposition are:—Bromination of the original acid in the α -position, conversion into α -hydroxy fatty acid, oxidation of the acid to aldehyde with lead tetra-acetate, and subsequent immediate oxidation of the aldehyde to the required lower fatty acid with air and excess of lead tetra-acetate. Nearly quantitative yields (*i.e.* up to 96 per cent. of the theoretical) of the hydroxy fatty acids were obtained. The yields of lower fatty acids were 88 to 89 per cent. of the theoretical calculated on the hydroxy fatty acid, or approximately 84 per cent. on the original fatty acid. The original describes the decomposition in detail and discusses results obtained.

E. B. D.

Reactions of Nessler's Reagent with Aqueous Solutions of Mustard Gas and Lewisite. J. Delga. (*J. Pharm. Chim.*, 1940, 1, 5–8.)—It has been observed that Nessler's reagent when added to water contaminated with dichlorodiethyl sulphide or with the mixture of chlorovinyl chloroarsines and trichlorovinyl arsine gives reactions likely to cause error in the normal use of the reagent or, conversely, reactions available for the detection of these toxic substances in water. With mustard gas the reagent gives a white precipitate that becomes yellowish if the concentration of mustard gas is sufficiently high. A study of the composition of this compound is in progress. The reaction is given with as little as 0.078 g. of mustard gas per litre. Thiodiglycol (formed by hydrolysis of mustard gas) gives no precipitate with Nessler's reagent and only a faint yellow colour when its concentration exceeds 6 g. per litre; it is therefore not responsible for the reaction. With Lewisite the reaction varies with the concentration and the time of reaction. The relation between the reaction and the concentration (expressed as g. of arsenic per litre) may be summarised as follows:—2.93 to 1.47, a white precipitate becoming grey in 2 minutes; 0.293, a chestnut-coloured precipitate becoming grey; 0.147 to 0.0293, an orange-yellow to yellow colour becoming chestnut and followed by a grey precipitate; 0.0097, a clear pale yellow colour changing to rose in 3 minutes; 0.0029, a faint greenish-yellow colour becoming more intense. The reaction is still perceptible at a concentration of 0.001 g. of arsenic per litre. The arsenic was determined by the method of Fleury (*J. Pharm. Chim.*, 1920, 21, 385; *Abst.*, *ANALYST*, 1920, 45, 389) after ignition. The solutions were prepared from the commercial liquid and were filtered and diluted until free from undissolved globules. The best results are obtained by adding 2 ml. of Nessler's reagent, drop by drop, with constant shaking, to 10 ml. of the solution.

A. O. J.

Precipitation of Natural Tannins with Calcium Hydroxide. V. Nemec. (*J. Inst. Leather Trades Chem.*, 1940, 24, 5–8.)—Calcium hydroxide in the form of a fine dry powder is added in excess (2 g. per 50 ml.) to the analytical tannin solution, and the mixture is boiled for 5 minutes beneath a reflux condenser, then cooled and filtered, and 25 ml. of the filtrate are evaporated to dryness in a quartz or platinum basin. The residue is weighed and then ignited in an electric oven at a

moderate temperature until a white ash is obtained. Errors due to partial conversion of calcium oxide into carbonate by atmospheric carbon dioxide are eliminated by adding a few drops of dilute ammonium carbonate solution to the cooled ash, which is then dried for one hour at 170° C. before weighing. In examining the portion not precipitated by calcium hydroxide the colours of the filtrates and dry residues are noted (*e.g.* valonea, colourless; sumach, yellow; quebracho, brown). The following amounts of tannin were thus determined:—*Hydrolysable tannins*.—Valonea, 68·9; myrobalans, 65·2; sumach, 25·4; oak wood (mean of 5 determinations), 66·8; chestnut wood (mean of 5), 70·3. *Condensed tannins*.—Ordinary quebracho, 69·5; sulphited quebracho, 73·3; mangrove, 65·6; mimosa, 69·3; pine bark, 65·0. With the hydrolysable tannins the portion not precipitated by calcium hydroxide is always smaller than the quantity of non-tans, as estimated by the hide-powder method; probably part of the sugar is precipitated. With the condensed tannins, however, sugar is not precipitated with the tannin.

D. G. H.

Absorption of Substantive Dyes by Oxy-Celluloses of the Acidic Type. S. M. Neale and W. A. Stringfellow. (*J. Soc. Dyers and Col.*, 1940, 56, 17–18.)—Cotton that has been oxidised by alkaline oxidising agents exhibits weakly acidic properties, presumably due to carboxyl groups. It shows an enhanced affinity for basic dyes, *e.g.* methylene blue, and a much reduced power of absorbing substantive dyes. In the experiments described a good grade of bleached Egyptian cotton cloth was used, the oxidising agent being a 0·1 *N* solution of potassium hypobromite in free alkali; dyeing was carried out at 90° C. and *pH* 6·20, with solutions containing 0·05 g. of dye and 5·0 g. of sodium chloride per litre. It was found that the amount of Sky Blue-FF absorbed fell off rapidly as the carboxyl value (determined by the authors' method, *Trans. Faraday Soc.*, 1937, 33, 881) increased, until a constant value was reached. The theory underlying these findings is discussed in detail.

J. G.

Precipitation Reactions of Organic Arsenic Compounds. M. Péronnet and R. H. Rémy. (*J. Pharm. Chim.*, 1939, 30, 353–364.)—About 25 organic arsenic compounds were tested. *Reaction with hydrogen sulphide*.—To the solution of the compound in aqueous alcohol (10 per cent. alcohol) was added 1 ml. of hydrogen sulphide water. A yellow precipitate or turbidity formed immediately with the majority of the compounds. The precipitate obtained with phenyl-dichloroarsine and methyl phenylarsonate was identified as phenylarsine sulphide and that with β -chlorovinyl-dichloroarsine as chlorovinylarsine sulphide. It is noteworthy that arsinic and arsonic acids gave no precipitate. The reaction obtained in aqueous alcoholic solution did not always occur when absolute alcohol was used as solvent. Hydrogen sulphide in acetone solution gave similar results to hydrogen sulphide water. *Reaction with mercury reagent*.—The reagent contained 2 ml. of nitric acid (sp.gr. 1·42) and 10 g. of mercuric nitrate in 100 ml. of water. A white or yellowish precipitate of unknown composition was obtained with most organic arsenic compounds in absolute alcoholic solution. Exceptions were compounds in which several aliphatic or aromatic groups were linked to arsenic in open chain.

S. G. C.

Inorganic

Reaction of Copper with Benzidine. G. Spacu and C. G. Macarovici. (Abstr., *J. Pharm. Belgique*, 1939, **21**, 1005; cf. *Chem. Abstr.*, 1939, **33**, 61944.)—The effect of anions on the reaction is dealt with. In presence of iodide, a complex cupric benzidine iodide is formed yielding a blue colour at a dilution of 1 in 2,000,000. In absence of copper, benzidine and iodide give an evanescent blue colour at a dilution of 1 in 1,000,000. In presence of halogens the sensitiveness of the copper-benzidine reaction decreases in the order iodide, bromide, chloride. Fluoride and cyanide prevent the reaction. S. G. C.

Tests for Cadmium and Magnesium. E. Eegriwe. (*Z. anal. Chem.*, 1939, **118**, 98–100.)—*Cadmium*.—When a cadmium salt solution is treated with sodium carbonate solution and a certain quantity of a solution of diaminoazobenzene in acetone, an orange-red turbidity or precipitate is formed, which, when shaken with chloroform, is extracted and forms an orange-red to red solution. Most of the other cations give only a yellow turbidity or precipitate, colouring the chloroform slightly yellow. This test, which is very sensitive, is carried out as follows:—Add to one drop of the neutral solution to be tested (free from copper and ammonium) 0.5 ml. of sodium carbonate solution (15 g. of $\text{Na}_2\text{CO}_3 \cdot 2\text{H}_2\text{O}$ in water made up to 100 ml.), 0.25 ml. of the reagent solution (0.5 g. of diaminoazobenzene in 100 ml. of acetone), and a few drops of chloroform and shake. In presence of 0.2% of cadmium the chloroform becomes orange-red. With copper the reagent gives a greenish colour, extracted by chloroform; silver causes a brownish-yellow turbidity in the chloroform layer. Both cations interfere with the reaction, as do also coloured precipitates formed by sodium carbonate or large proportions of cations giving colourless precipitates; also ammonium salts. Reactions similar to that with cadmium are given by cobaltous salts and by nickel. Whereas, however, the cobalt colour disappears on shaking, whilst the red colour of the chloroform changes to brown, the nickel colour remains unchanged. Thus cadmium can be detected in presence of cobalt; also, nickel in presence of cobalt. For the detection of cadmium in presence of small amounts of nickel, solid dimethylglyoxime is added to remove the nickel from the chloroform layer, and after addition of the reagent an orange-red colour then indicates the presence of cadmium. By this method 3% of cadmium can be detected in presence of ten times the amount of nickel. The limit for the identification of nickel is 0.05% of nickel in 1 drop of the solution; 3% of cadmium can be detected in presence of 500 times the amount of lead, manganese(ous) and zinc, but not of mercury(ous), bismuth, aluminium, calcium or magnesium; in presence of only 100 times the amount of these cations, however, the detection of cadmium is possible.

Magnesium.—On adding to 1 drop of pure water in a test-tube a few drops of conc. ammonia, then a small amount of solid *p*-aminophenol hydrochloride, shaking a few times and allowing the tube to stand, the liquid is gradually coloured by oxidation from faint yellow, through yellow-brown, to brownish. In presence of magnesium the oxidation product is adsorbed by the magnesium hydroxide, and instead of the yellow and brown colour a light to deep blue colour appears.

This reaction has not been observed with any other cation. In applying the test 1 drop of the neutral solution of the fifth group, free from ammonium salts, is used. Cations forming precipitates with ammonia, or anions giving less soluble magnesium components (phosphate or arsenate ions), interfere with the test; also sulphites, which prevent the necessary oxidation of the reagent. The blue colour, or the formation of blue flakes, is still visible with 5γ of magnesium in 1 drop (0.05 ml.) of solution. With smaller amounts of magnesium, or in presence of cations that interfere with the reaction (*e.g.* cadmium and zinc), no blue colour appears, but the solution shows an initial violet colour when the normal amount of reagent is used. Excess of the reagent gives a violet colour also with magnesium; the violet colour may be obtained with 1γ of magnesium. The characteristic blue colour may also be used as a macro-reaction. D. A.

New Method for the Volumetric Determination of Bismuth. L. Malaprade. (*Ann. Chim. anal.*, 1940, 22, 5-8.)—The method involves (a) neutralisation of free acid in presence of an excess of sodium thiosulphate, yielding a complex sodium bismuthi-thiosulphate which is stable at pH 4; (b) titration of the complex bismuth salt with alkali according to the reaction



(a) To the solution in dilute nitric acid (100 ml.) containing 0.17 to 0.5 g. of bismuth, 4 drops of methyl red indicator solution (2 per cent. in alcohol) are added, and potassium hydroxide solution is introduced until the indicator changes to yellow and a precipitate of basic bismuth salt is produced; 10 g. of sodium thiosulphate are dissolved in the solution. Dilute nitric acid is added, drop by drop, until the precipitate re-dissolves, and the solution is then neutralised with a few drops of potassium hydroxide solution, as necessary, yielding a pale orange colour (faint red colour of the methyl red superposed on the yellow colour of the bismuth complex). (b) Ten drops of phenolphthalein indicator (2 per cent. in alcohol) are added, and the solution is titrated with 0.2 N potassium hydroxide solution. The colour change at the end-point is from yellow to a neutral tint produced by the superposition of the yellow colour of the liquid on the faint pink of the phenolphthalein. It was established that, at the end-point, 1 ml. of 0.2 N potassium hydroxide solution is equivalent to 16.72 mg. of bismuth. The solution titrated should be free from chloride and sulphate, which interfere with the reaction in (b). The alkali solutions used should be as free as possible from carbonate. Bismuth may be determined after separation as sulphide. The sulphide precipitate is dissolved in conc. nitric acid, the liquid is diluted, the sulphur is filtered off, and a slight excess of barium nitrate is added to precipitate sulphate formed by oxidation of the sulphide. The barium nitrate and sulphate present are without effect on the subsequent determination of bismuth, which is carried out as described above. S. G. C.

Colorimetric Determination of Ferric Iron by means of Gallic Acid. Y. Volmar and A. Wagner. (*J. Pharm. Chim.*, 1939, 30, 364-369.)—The method is applicable to 1 to 6 mg. of ferric iron. The solution in dilute hydrochloric acid is evaporated to remove excess acid. The residue is dissolved in 5 ml. of water,

3 ml. of gallic acid solution (2 per cent.) are added, and the solution is diluted to 15 ml. with saturated sodium acetate solution. The bluish-violet colour produced is compared colorimetrically in a Duboscq colorimeter with a similarly prepared standard containing 2 mg. of ferric iron. The method may be applied to the determination of iron in blood: 3 to 5 g. of blood are rendered alkaline with ammonia and evaporated to dryness. The residue is mixed with 5 to 15 g. of ammonium nitrate, and the mixture is heated to destroy organic matter and finally ashed. The ash is dissolved in 2 to 3 ml. of conc. hydrochloric acid with the addition of a little hydrogen peroxide, the liquid is evaporated to remove excess acid, and the iron is determined as described above. S. G. C.

Determination of Chlorides, Thiocyanates and Cyanides in Presence of each other, and a Qualitative Reaction for the Detection of Chlorides in Potassium Thiocyanate. A. Slooff and D. Van Duyn. (*Chem. Weekblad*, 1940, 37, 69-72.)—In Treadwell's method (*Lehrb. anal. Chem.*, 1927, II, 621) the chlorides, thiocyanates and cyanides are titrated together by Volhard's method and the cyanides only by Liebig's method, the thiocyanates plus cyanides being determined by boiling a mixture of the three silver salts with nitric acid, and titrating the soluble portion with a standard ammonium thiocyanate solution. This assumes, although unjustifiably, that no silver chloride dissolves; even if this error is minimised by the use of less concentrated nitric acid, the decomposition of the silver cyanide and thiocyanate is slow and incomplete. In the present method the Volhard and Liebig titrations are used as in Treadwell's method, and the chlorides are then isolated by destruction of the cyanides and thiocyanates with hot nitric acid. Thus, a mixture of 100 ml. of the solution and 10 ml. of nitric acid (sp.gr. 1.3) is heated slowly to boiling so that the rapid oxidation of the cyanides and thiocyanates does not cause it to boil over; if thiocyanates are present the solution is dark red, but subsequently becomes colourless. The mixture is boiled gently for 30 minutes, precautions being taken to avoid bumping, an excess of 0.1 *N* silver nitrate solution is added, and, after further boiling for a short time (to coagulate the silver chloride), the solution is cooled, and the excess of silver nitrate is back-titrated with 0.1 *N* ammonium thiocyanate solution, with ferric ammonium alum as indicator. Sulphates produced by oxidation of the thiocyanates do not interfere. With 24.8 to 124.0 mg. of chloride, in presence of 58.4 to 292.0 mg. of thiocyanate and 51.3 to 256.5 mg. of cyanide, the differences between the amounts of chloride taken and found ranged from +0.2 to -0.4 mg. In the Liebig titration high values were found for the cyanide-content when the thiocyanate and chloride contents were high (*e.g.* above about 60 mg. per litre), and it is therefore desirable first to dilute such solutions. It was found that in the determination of small quantities of chlorides in potassium thiocyanate the authors' method gave results agreeing well with those obtained by difference from a determination of the thiocyanate content by Volhard's method or by Rupp's method (*i.e.* oxidation with iodine in presence of alkali, and back-titration of the excess of iodine). Rivot's method (precipitation of the thiocyanate with copper sulphate), or oxidation of the thiocyanate by means of bromine water to sulphate, which is determined as barium sulphate (*cf.* Treadwell, *loc. cit.*, pp. 290, 291) gave

low results for the thiocyanate-content. In the qualitative test for chlorides in potassium thiocyanate a solution of 1 g. of the sample in 100 ml. of water is boiled for 30 minutes with 10 ml. of nitric acid (sp.gr. 1.3), silver nitrate solution being added to the warm mixture so as to avoid the precipitation of silver sulphate; an opalescence indicates the presence of chlorides. The authors collected in sodium hydroxide solution the gases evolved from a mixture of 1 g. of potassium thiocyanate and 10 mg. of potassium chloride during the boiling process; they demonstrated by the above method that under these conditions no chlorides are lost by volatilisation.

J. G.

Microchemical

Detection of Barium and Sulphate by means of Spot Reactions. F. Feigl and W. Aufricht. (*Rec. Trav. Chim. Pays-Bas*, 1939, **58**, 1127–1132.)—Potassium permanganate which has been adsorbed from solution by barium sulphate adheres to it permanently and the resulting violet colour of the sulphate is almost unaffected by the usual permanganate-reducing agents (*cf.* Wohlers, *Z. anorg. Chem.*, 1908, **59**, 203). A spot method based on this effect has been devised for the detection of small amounts of barium and sulphate. To obtain maximum visibility, it is essential to precipitate the barium sulphate in concentrated permanganate solution and to reduce the excess of permanganate immediately without filtering. For the detection of barium, sulphurous acid is the best reducing agent (as the sulphate formed decreases the solubility of barium sulphate). The test can be made with one drop of the test solution on the spot plate, in Emich's small pointed tubes, or on filter-paper—the limiting sensitivities for barium by these methods being 12, 2.5 and 5 γ , respectively, and the corresponding limiting concentrations, 1 to 4200, 20,000 and to 10,000.

Detection of Barium.—(a) *Spot plate.*—One drop (0.05 ml.) of the test solution and 3 drops of cold saturated potassium permanganate solution are mixed on the plate, the barium is precipitated with a few drops of dilute sulphuric acid, and sulphurous acid is added immediately while the liquid is stirred with a sealed glass capillary tube until the solution is decolorised. A pink turbidity indicates barium. (b) *Emich tube.*—The reaction is carried out as before. After decolorisation of the solution the barium sulphate is centrifuged into the tip of the tube, where it can readily be recognised against a white background, particularly by means of a lens. (c) *Filter-paper.*—A sulphate paper which can be stored after drying is prepared by soaking strips of filter-paper in $N/2$ sodium sulphate solution. One drop of a mixture of 3 drops of the test solution with one drop of the permanganate solution is placed on the dry sodium sulphate paper, which is then kept for 7 to 10 minutes in an oven at 70° C. to 80° C. The original violet colour changes to brown, owing to reduction of the permanganate by the filter-paper; longer drying weakens the sensitivity of the reaction. The dried paper is soaked in sulphurous acid for 1 or 2 minutes, until the last traces of manganese dioxide have been removed. The presence of barium sulphate on the paper, which is now white, is shown by a violet fleck or ring. Under the same experimental conditions strontium sulphate is very slightly coloured by potassium permanganate solution,

whilst calcium sulphate does not adsorb permanganate. It is possible to detect 5 γ of barium in presence of 2500 γ of strontium if a blank test with pure strontium nitrate is made. While decolorisation of the coloured barium sulphate by conc. sulphurous acid is very slight, even in 24 hours, that of the coloured strontium sulphate is rapid and complete. *Detection of Sulphate.*—(i) *Spot method.*—To a mixture of one drop of the test solution and three drops of cold saturated permanganate solution on the spot plate is added 1 drop of $M/2$ barium chloride solution and, after mixing, 3 to 4 drops of 3 per cent. hydrogen peroxide to decolorise the excess of permanganate. The barium sulphate is coloured violet. For the better recognition of the colour in the spot-plate tests for sulphate and for barium it is advisable to fill an adjacent depression in the plate with water. Limiting sensitivity: 3 γ of sulphuric acid; limiting concentration: 1 to 9000. (ii) *Emich tube.*—The test is made as in (i), hydroxylamine hydrochloride being used as reducing agent. The centrifuging and examination of the precipitate are as in (b). Limiting sensitivity: 2.5 γ of sulphuric acid; limiting concentration: 1 to 20,000. (iii) *Filter-paper.*—One drop of a mixture (3:1) of the test solution and of saturated permanganate solution is placed on filter-paper which has been soaked in $M/2$ barium chloride solution and dried. After drying again at 70 to 80° C. in an oven for 7 to 10 minutes the excess of barium chloride is removed by washing out with water in a dish for one minute, then placing for an instant under the water-tap. On placing the paper in oxalic acid solution a pink ring indicates the presence of sulphate. Limiting sensitivity: 2.5 γ of sulphuric acid; limiting concentration: 1 to 20,000. Lead sulphate gives a similar reaction under these conditions. The method for the detection of lead will be given later.

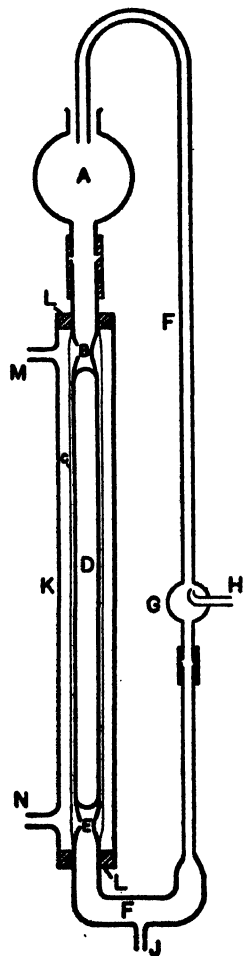
E. B. D.

Microscopy of the Amino Acids and their Compounds. IV. Picrolonates.

R. Dunn, K. Inouye and P. L. Kirk. (*Mikrochem.*, 1939, 27, 154–160.)—Picrolonates were obtained from all naturally occurring amino acids and from some synthetic amino acids not found naturally. Proline yields a salt only from concentrated solution. A minute amount of the amino acid added to the solution of about 0.01 mg. of the reagent produces satisfactory crystals with alanine, arginine, cysteine, dibromotyrosine, dichlorotyrosine, glycine, histidine, norleucine, norvaline, tyrosine and valine. The remaining amino acids require a more concentrated solution to produce satisfactory crystals. The predominant crystal habit is acicular, the needles being arranged in rosettes; the appearance is insufficiently characteristic, but in nearly every instance the refractive indices serve for identification. These are measured in the two extinction positions 90° apart, designated N_1 and N_2 . The values obtained are as follows: alanine, $N_1 = 1.575$, $N_2 = 1.580$; arginine, (a) $N_1 = 1.716$, $N_2 = 1.580$, (b) $N = 1.578$; aspartic acid, (a) $N_1 < 1.527$, $1.527 > N_2 > 1.512$, (b) N_1 and $N_2 > 1.740$; cysteine, N_1 and $N_2 > 1.740$; cystine, $N_1 = 1.600$, $N_2 = 1.548$; dibromo-tyrosine, (a) $N_1 > 1.740$, $1.616 > N_2 > 1.549$, (b) $N_1 < 1.549$, $N_2 > 1.580$; dichloro-tyrosine, $1.740 > N_1 > 1.698$, $1.633 > N_2 > 1.618$; diiodo-tyrosine, anisotropic; N_1 and N_2 could not be measured; glutamic acid, $N_1 = 1.574$, $N_2 = 1.596$; glycine, $N_1 = 1.616$, $N_2 = 1.531$; histidine, $N_1 = 1.616$, $N_2 = 1.557$; hydroxyproline,

$N_1 = 1.658$, $N_2 = 1.493$; hydroxy-valine, $N_1 = 1.56$ (approx.), $N_2 = 1.54$ (approx.); isoleucine, $N_1 = 1.610$, $N_2 = 1.520$; isoserine, (a) $N_1 > 1.740$, $N_2 > 1.70$ (approx.), (b) $N_1 = 1.608$, $N_2 = 1.520$, (c) $N_1 = 1.660$, $N_2 = 1.529$; leucine, $N_1 = 1.617$, $N_2 = 1.527$; lysine, $N_1 = 1.645$, $N_2 = 1.520$; methionine, $N_1 = 1.62$ (approx.), $N_2 = 1.494$; norleucine, $1.740 > N_1 > 1.658$, $1.740 > N_2 > 1.658$; norvaline, (b) $N_1 = 1.684$, $N_2 > 1.74$; phenylalanine, (b) N_1 and $N_2 > 1.74$; proline, $N_1 = 1.530$, $N_2 = 1.605$; serine, (a) $N_1 = 1.567$, $N_2 = 1.530$, (b) refractive indices not measured owing to rapid solution in immersion media; tryptophane, $N = 1.712$, tyrosine, $N_1 = 1.596$, $N_2 = 1.529$; valine, $N_1 = 1.549$, $N_2 > 1.740$; *dl*- α -amino-*n*-valeric acid, $N_1 > 1.685$, $N_2 > 1.740$. Twelve photomicrographs are given. The letters (a) and (b) represent two types of crystals. J. W. M.

Physical Methods, Apparatus, etc.



A Rapid Circulating Dialyser. A. R. Taylor, A. K. Parpart and R. Ballentine. (*Ind. Eng. Chem. Anal. Ed.*, 1939, 11, 659.)—In this apparatus a current of water flows from M to N through the outer glass jacket K. The dialysing membrane C consists of a thin sheet of cellophane, and a glass cylinder D, which is enclosed in the membrane and kept in position by fusion with the inlet and outlet tubes B and E, ensures that the liquid under treatment is in a thin layer. The outlet J is closed with a rubber tube and clip, and the liquid to be dialysed is poured into the bulb A, whence it descends, filling the lower part of the apparatus and part of the tube F. A current of air or inert gas, passed through the jet H, causes the solution to circulate, and samples can be drawn off at J for examination. As an example of the efficiency of the apparatus it is stated that 25 to 100 ml. of protein solution semi-saturated with ammonium sulphate were completely freed from the salt in nine hours. Another advantage claimed for the apparatus is that there is no risk of rupturing the cellophane membrane, which can be used repeatedly.

Reviews

MICRO-DIFFUSION ANALYSIS AND VOLUMETRIC ERROR. By EDWARD J. CONWAY, M.B., D.Sc. Pp. xiii + 306. London: Crosby Lockwood & Son, Ltd. 1939. Price 25s. net.

The micro-diffusion method of analysis is so simple and the apparatus so cheap that it deserves a much wider application than as at present mainly in the problems of medical and biochemical research. It is to be hoped that Professor Conway's excellent book, which includes an account not only of the method but also of micro-volumetric procedure and the errors of volumetric determination, may achieve this result by interesting more analytical chemists in the method.

The standard micro-diffusion unit resembles two small concentric Petri dishes, the walls of the internal portion being half the height of the outer. When sealed with an airtight cover this simple apparatus may be used for the determination of volatile gases liberated by a reagent in the outer chamber, absorbed by a reagent in the centre chamber, and subsequently measured by titration or colorimetrically.

Three chapters are devoted to the use of the unit itself, the standard procedure and generalised conditions for the determination of amounts of test substance (ammonia, urea, chloride, bromide and carbon dioxide) ranging from 0.5 to 500 γ per ml. (designated "gammis," by the author) in 1 ml. volume, with an accuracy of 0.5 per cent. for amounts of 20 γ and more and 5 per cent. for 0.4 "gammil."

The chapters on volumetric procedure are extremely valuable. The greater accuracy, well known to micro-chemists, of using simple laboratory-made pipettes, is clearly explained. The simple tube-shape pipette is shown to be quite as accurate as the original Ostwald form, if not more so.

In the second part of the book the different methods are described in detail, in thirteen chapters, the final chapter being devoted to Karon and Webb's qualitative tests for acetone and alcohol which were described in this journal three years ago.

The chapters on error are readily understandable without any very great knowledge of mathematics, and are of great value to all those to whom accuracy is important, that is, to all analysts.

The work can be highly recommended not only to those interested in diffusion analysis, but to all who wish to have a clear exposition of micro-volumetric procedure, including "drop" technique, with especial reference to the accuracy of the results. This is the best book on micro-volumetric work that the reviewer has had the pleasure of reading.

JANET W. MATTHEWS

A LABORATORY MANUAL OF QUALITATIVE ANALYSIS. By JOHN H. YOE, Ph.D. Pp. ix + 219. London: Chapman & Hall, Ltd. 1938. Price 12s. 6d. net.

In this text-book of elementary inorganic qualitative analysis, intended to cover a thirty-six weeks' course, only such reactions of cations and anions are included as are used for their identification in the schemes for separation. The space saved by the exclusion of the general reactions of the elements has been

devoted to a very complete description of the procedures used for their separation and detection and for detailed explanatory notes following each section.

Intended to be used solely as a bench book, it contains no physico-chemical theory; for this the student is referred to the author's or to other works on the subject. This should please those who consider that the art and the science of analytical chemistry are best treated separately; such a thought was probably in the mind of the author when he wrote, page 42, "Qualitative analysis was a well organised art long before Wilhelm Ostwald placed it upon a firm scientific basis when he published his '*Wissenschaftliche Grundlagen der analytischen Chemie*,' 1894."

The work covers the usual twenty-four common cations of the elementary course, and the same number of anions; it follows American practice in precipitating the hydroxide and alkaline sulphide groups together. An unusual feature is the scheme for the separation of arsenic, antimony and tin by successive precipitation, under controlled conditions of acidity, as sulphides from hydrochloric acid solution. The reviewer, having in mind the well-known adsorptive capacity of antimony sulphide for tin, was agreeably surprised to learn that one milligram of tin in a one per cent. mixture with antimony gave, under the specified conditions and after filtration, a characteristic and easily visible precipitate of stannic sulphide.

As a text-book it may be considered defective by many in this country in that it contains no provision for elimination of phosphates or other interfering elements in the iron group, and it is somewhat diluted by the inclusion of standing orders to the students of a particular school of chemistry.

A special feature is a section on delicate reactions by means of organic reagents, to which reference is made at appropriate points in the text. In this, possibly the most valuable part of the work, the manipulation is fully described, with warnings as to interfering elements and references to original sources; the reactions, wherever possible, are illustrated by structural formulae.

One misprint was noted on page 167, where "Janus Green" is termed "James Green."

Although there are elementary text-books of wider scope at more moderate price than this, there are but few in which the subject-matter is presented with more detailed precision.

F. L. OKELL

HANDBOOK OF FOOD MANUFACTURE. By F. FIENE and S. BLUMENTHAL, B.Sc.
Pp. vi + 603. London: Chapman & Hall. 1939. Price 25s. net.

This ambitious attempt to provide information of service to the various classes of workers engaged in the manufacture of foods and beverages deals mainly with the practical aspects of the industry, from the sources, grading and testing of the raw materials to the preparation of the finished products. The profusion of formulae and methods described are based upon large-scale industrial experience, and thus may be relied upon to produce successful results.

Most of the extensive range of materials dealt with would be included under the headings of milk, meat, fish, fruit and nut preparations, pâtisserie, confectionery, preserves, sauces, liqueurs, cocktails, and fruit drinks, while mixtures sold to the public for the home preparation of pastry, ices and so forth are also described.

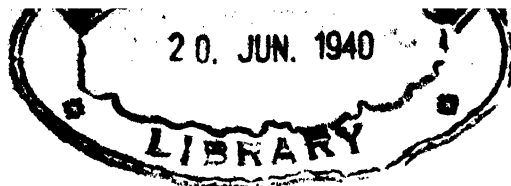
The first part of the chapter entitled "An Outline of Elementary Chemistry" is too condensed to be of value either to the layman or the chemist and might well have been omitted; in contrast with this, the rest of the chapter contains admirable summaries of vitamin data and analytical values for the composition of many naturally occurring foods together with the physiological effects of the various metals, the halogens, phosphorus and sulphur in dietary. Over sixty pages are devoted to the numerous raw materials used in the food industry, including *inter alia* synthetic compounds, essential and fixed oils, drugs and cereals, the physical properties and taste effects of many of these being given in detail. This chapter is excellent, but it is a little disconcerting to find in a book on foods that human, horse, dog and wool fats are included in a table giving the solidifying points of fats (p. 68), and the value of 9 per cent. given for the yield of sucrose from the sugar-beet is much below the amount obtained at the present time.

The following twenty-three chapters contain a miscellaneous collection of formulae and procedures for the manufacture of hundreds of food products, the variety of which would overwhelm the famous Mrs. Beeton. This section is undoubtedly the best in the volume. Chapter XXV provides an admirable account of hydrogen ion control by means of indicators and its applications, *e.g.* in bread-making and canning. Among the "Miscellaneous" items is a useful section on the use of alkalis for bottle-washing together with the requirements of many of the American States in this respect. The striking diversity in the concentrations of alkali (2.5 to 5 per cent.), temperatures (100 to 160° F.), and time of soaking (3 to 20 minutes) causes one to wonder if such regulations have any value, especially as some States make no requirements whatever. One table giving the common adulterants of about sixty food materials will prove of considerable value to the analyst.

In the chapter dealing with "Analyses and Tests" a number of qualitative and quantitative tests for a wide range of food materials are given. Many of these are satisfactory, but the precautions necessary for the correct interpretation of results obtained in the colour-tests for oils are omitted, and no reliance can be placed upon the results of some of the tests as described. It is evident that the proofs were not read by a qualified chemist, otherwise numerous erroneous statements in this section would have been eliminated.

The final pages of the volume contain the definitions and standards for food products adopted in the United States under the Food and Drugs Act, and an extensive and accurate index which, whilst not quite complete, yet contains nearly 2500 references. There is much sound and valuable matter in the text, but its merits are greatly discounted by the number of errors present, and it may be hoped that these will be eliminated by more thorough proof-reading in the next edition.

T. J. WARD



THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

AN Ordinary Meeting of the Society was held at 5.30 p.m. on Wednesday, April 3rd, 1940, at the Chemical Society's Rooms, Burlington House, the President, Dr. E. B. Hughes, in the chair.

Nominations were read in favour of Sir William Willcox, K.C.I.E., C.B., C.M.G., M.D., F.I.C., proposed by the Council for Honorary Membership, and the following applicants for Ordinary Membership:—J. A. Freeman, B.Pharm., B.Sc., F.I.C., Ph.C., M.P.S., and J. T. Stock, B.Sc.

The following papers were read and discussed:—"Hair Dyes. Part II. The Functions and Reactions of Phenols," illustrated by a cine-film in colour showing the process of hair-dyeing, by H. E. Cox, D.Sc., Ph.D., F.I.C., and "The Determination of Aluminium, Magnesium or Beryllium in Nickel Alloys," by R. C. Chirnside, F.I.C., L. A. Dauncey, B.Sc., and P. M. C. Proffitt.

THE SAUSAGES (MAXIMUM PRICES) ORDER, 1940 STATUTORY RULES AND ORDERS, 1940. NO. 394

The Council has adopted the following recommendation of the Analytical Methods Committee of the Society.

For the purposes of the above Order "Meat Content" should be determined as follows:—

The process consists in the chemical determination of water, protein (nitrogen $\times 6.25$), fat and ash. The calculation of the meat content is made on the lines advocated by Stubbs and More (ANALYST, 1919, 44, 125), with the exception that the factors for converting meat nitrogen into de-fatted meat should be 100/3.4 for beef or mutton, 100/3.6 for pork.

Detailed methods of analysis are given in the above-mentioned paper of Stubbs and More. An alternative method for the determination of fat may be employed, comprising digestion of 2 to 3 grams of the sample with about 20 millilitres of hydrochloric acid (1 volume of strong hydrochloric acid diluted with from 1 to 2 volumes of water) at a temperature just below boiling-point until the meat fibre is disintegrated and all the fat is liberated, with subsequent extraction of the fat with ether.

Obituary

SIR WILLIAM JACKSON POPE, K.B.E., F.R.S.

THE death of Sir William Jackson Pope, at the age of 69, inflicted a loss felt far beyond the bounds of his own country, for he had won a great position in the world of chemistry.

He was an outstanding product of the system of technical education established in this country in the latter part of the last century; technical colleges provided his chemical training, they gave him his earlier teaching appointments, and in them he laid the foundations of his great scientific reputation.

He was born in North London in 1870, the eldest of eight children. His parents were William Pope, a native of Biggleswade, and Alice Hall, of Prudhoe, Northumberland. They were married in 1869.

One of the earliest students at the Finsbury Technical College, he came at once under the stimulating influence of H. E. Armstrong, and when Armstrong was made Professor of Chemistry at the Central Technical College, Pope went with him and later became his assistant. A strong attachment existed between the two men; Armstrong followed with unbounded pride the career of his most distinguished pupil, and Pope retained an almost filial regard for his one-time teacher.

In 1897, at the age of 27, he received his first appointment, the Headship of the Chemical Department of the Goldsmiths' Company's Institute at New Cross—he was afterwards to become Prime Warden of the Company—and, four years later, left it to become Professor of Chemistry and Head of the Chemical Department at the new Manchester Municipal School of Technology, where he remained for seven years.

His election to the Professorship of Chemistry at Cambridge took place in 1908, on the retirement of Professor G. D. Liveing, and in the following year he was elected to a professorial fellowship at Sidney Sussex College.

During the world war he served as a member of the panel of consultants of Lord Fisher's Board of Invention and Research, and his energies were directed unsparingly towards finding solutions of the chemical problems with which the country was then faced. The strong practical sense with which his great knowledge was combined made his help particularly effective and, in connection especially with the sources of high explosives, aerial photography, and retaliation to the German use of poison gas, he rendered assistance of the highest value. His services received recognition in the K.B.E. conferred on him in 1919.

After the war Pope became more and more involved in the administration of chemical affairs, a sphere in which his keen and subtle mind inevitably brought him to the front. In 1918, in his presidential address to the Chemical Society, he had advocated the formation of a body to co-ordinate the activities of the societies representing the different chemical interests in this country. Largely through his efforts the body thus foreshadowed was soon afterwards brought into being as the Federal Council for Pure and Applied Chemistry, and he was its first chairman. At the same time he was engaged, with some of the more prominent

chemists of the allied countries, in bringing about the formation of the *Union Internationale de Chimie*, designed to co-ordinate in a somewhat similar fashion the chemical societies of different countries. From 1922 to 1925 he served as president of this organisation, and in June, 1923, the Fourth International Chemical Conference, attended by a distinguished gathering of British and foreign chemists, met in Cambridge under his leadership.

Another mark of the esteem in which he was held by his Continental colleagues was his selection to preside—during the period 1922 to 1936—over the Chemical Conferences held in Brussels under the Solvay Foundation. At these assemblies of the foremost authorities on the subject to be debated his linguistic powers were much in evidence, for during the discussions he had frequently to act as interpreter, translating German contributions into French or French into German, and he made admirable after-dinner speeches in French at the banquets with which the conferences concluded. In appreciation of his services he was, in 1937, created Grand Officier de l'Ordre de Léopold. Many medals, including the Davy Medal of the Royal Society, were awarded him, he received many honorary degrees, and he was made a foreign member of learned societies of many lands.

His first original scientific communication was a joint paper with Armstrong, in which it was shown that pinene could be characterised through its oxidation-product, sobrerol. In this, his talent for crystallography was manifested. The crystallographic characters of dextro-, laevo- and racemic sobrerol were described, and the terpene fraction of American turpentine was shown to consist essentially of a mixture of *d*-pinene with a smaller and variable proportion of *l*-pinene.

Pope had studied crystallography under H. A. Miers and became deeply interested in it. In all his earlier researches much of the work was devoted to securing crystallographic data, and the hours he spent in the dark room with his goniometer were probably among the happiest in his life. These crystallographic studies had an important influence on the development of his chemical work, for they enhanced that natural faculty for visualising spatial relationships which drew him inevitably into the field of stereochemistry where his greatest achievements were won.

After the work on sobrerol was finished Armstrong asked him to examine the action of sulphuric acid on camphor in consequence of a communication by Marsh on this subject. Encountering experimental difficulties he sought the help of his colleague, F. S. Kipping, and thus began a scientific partnership which lasted several years and had an important effect on the future course of his work. The two investigators carried out an extensive examination of the sulphonation of camphor and its halogen derivatives, discovering the highly crystalline sulphonic derivatives of camphor and the series of halogenated camphors obtainable from them. They observed the phenomenon of *pseudo*-racemism, and among their other researches was a noteworthy investigation in which they showed that the addition of glucose or mannite to solutions of sodium chlorate (which crystallises in enantiomorphous forms) caused a remarkable disproportion between the numbers of *d*- and *l*-crystals deposited. The partnership was brought to an end by Kipping's departure to take up his professorship at Nottingham, and Pope shortly afterwards went to the Goldsmiths' Institute.

Here one of his first investigations was an attempt to resolve tetrahydro-papaverine in order to get evidence of the presence of the asymmetric carbon which Goldschmidt's papaverine formula would require. Tartaric acid, till then the only acid employed for resolving externally compensated bases, proved ineffective. He accordingly tried bromocamphorsulphonic acid, which he had got to know so well through his work with Kipping, and was immediately successful. Having got this indication of the efficacy of the camphorsulphonic acids—he attributed it to their strength and the exceptional crystallising power of most of their derivatives—he lost no time in applying them to one of the outstanding problems of the time—the resolution of asymmetrically substituted ammonium salts. Suitable material was already available in the benzyl-phenyl-allyl-methyl ammonium salts prepared, but found unresolvable by Wedekind. Pope, with Peachey, prepared the β -camphorsulphonate and, crystallising it from non-polar solvents, succeeded in effecting a resolution and obtaining the enantiomorphous optically active bromides and iodides. Thus for the first time an optically active compound owing its activity to an asymmetric atom other than carbon was prepared. This showed that the valencies of other elements besides carbon had sufficient configurational stability to give rise to observable optical activity when they were asymmetrically combined. It was a discovery which was at once recognised as one of first importance in stereochemistry, and it opened a wide field to investigation.

It was followed up by the resolution in rapid succession (with the collaboration of Peachey, Harvey and Neville) of asymmetric compounds of sulphur, selenium and tin. The sulphur compound examined was methyl ethyl thietine bromide (I), and this and the analogous methyl phenyl selenetine bromide (II)



were both resolved by means of bromo-camphor-sulphonic acid. Optically active tin was obtained by means of methyl ethyl propyl tin camphor- and bromo-camphor-sulphonates.

These investigations were made at a time when the distinction between electrovalency and co-valency was imperfectly understood, and the optically active compounds of quadrivalent sulphur, selenium and tin were regarded as formally analogous to compounds of asymmetric quadrivalent carbon. It is to be noted, however, that Pope expressly pointed out that they were salts and must be held to ionise, and that in the optically active ions three radicals only were attached to the asymmetric atom.

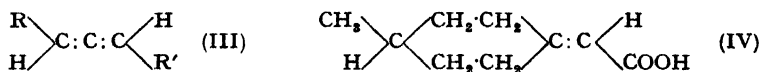
About the time that these investigations were completed he left New Cross for Manchester. His work at the Municipal School of Technology at first followed the lines he had pursued at the Goldsmiths' Institute. The experiments on selenium and tin were continued, as well as investigations of the stereochemistry of cyclic bases, but he soon broke new ground.

Using Grignard reagents, as for the alkyl tin halides, he prepared, with Peachey, trimethyl-platinic salts, and with Gibson, dialkyl gold halides, thereby showing for the first time that the noble metals were capable of combining with

organic radicals. With Read he began the investigation of asymmetric compounds of simple structure in order to determine what degree of molecular complexity was necessary to give rise to stable optical activity. Chloriodomethane sulphonic acid, with only a single carbon atom in the molecule, and chloriodo acetic acid with two, were eventually obtained in stable optically active forms, and many years later Read and McMath succeeded in finding a method for resolving chlorobromo-methane sulphonic acid.

At this time, also, he was engaged with W. Barlow in preparing the elaborate memoirs in which the Barlow-Pope valency-volume theory of crystal structure were expounded. Although the fundamental postulate of the proportionality between valency and volume cannot now be maintained, there was much in these memoirs—as, for example, the way in which the consequences of close-packing were developed—that foreshadowed present ideas based on X-ray analysis.

The outstanding investigation initiated in the Manchester period was, however, that on methylcyclohexylidene acetic acid. Pope conceived the brilliant idea of producing a compound which should be optically active and yet contain no asymmetric atom in its molecule—at least, none in the ordinary sense of the term. The molecular type which he devised as best suited for this purpose could be

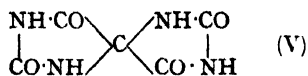


regarded as derived from a substituted allene (III) by the expansion of an ethylenic residue into cyclohexylidene, leading, with an appropriate choice of substituents, to the above-mentioned acid (IV).

For the production of this compound he secured the collaboration of W. H. Perkin, and a process for its synthesis was described in a joint paper in 1908. By a remarkable coincidence Marckwald and Meth were at the same time engaged on the synthesis of this compound for the same purpose. They prepared an acid which they believed to be methylcyclohexylidene acetic acid and resolved it into optically active components. It was, however, different from Perkin and Pope's acid. Since the latter was prepared by a process which definitely fixed its constitution, whilst Marckwald and Meth's synthesis allowed the alternative production of methylcyclohexene acetic acid, it was clear that Perkin and Pope's was the right compound, and Marckwald and Meth's the isomer with an ordinary asymmetric carbon atom. But although Perkin and Pope's process gave the right compound, it involved several difficult operations and could not easily be made to yield enough of the acid for resolution. Shortly afterwards, however, Wallach chanced upon a simpler synthesis, and the acid could then be readily obtained in quantity. The final realisation of Pope's idea through the resolution of the acid into optically active components (carried out with the assistance of Dr. John Read, now Professor of Chemistry at St. Andrews) was one of the first-fruits of his scientific work at Cambridge.

Of the results of the work of his later years, the most noteworthy were his contributions to our knowledge of the co-ordination compounds of aliphatic tri- and tetramines (with F. G. Mann) and the stereochemistry of spirocyclic compounds

(with J. B. Whitworth and S. E. Jansen). One of his last achievements, the resolution of *spirodihydantoin* (V)



(carried out with Whitworth) provided a particularly elegant demonstration of the molecular dissymmetry of compounds of this class.

As a lecturer Pope had an unusually facile delivery, and his lectures were remarkable for their clearness. Indeed, one felt there was some danger of his making organic chemistry seem so easy that the more able students might underestimate the serious study it required. He directed his department with little apparent effort. Exerting a minimum of interference, but giving constant support and encouragement, he knew how to get the best out of his staff.

Despite an air of apparent gloom he could be a most entertaining companion, for he had a rich fund of delightful stories. They were often based on simple incidents of the laboratory or his earlier experiences in London, but he told them inimitably, and they almost invariably had an exquisitely humorous climax.

To those who did not know him well he might seem not readily approachable, but this apparent reserve concealed deep stores of kindness. His help was ever forthcoming to those who were in any way in real need of assistance, and he did many acts of kindness that were scarcely known except to himself and the recipient.

He was a gracious host, and a visit to his house was always full of interest. He had an unusually fine chemical library, and his collections of pharmaceutical jars and mortars and pestles are probably unique. He had also a fine collection of alchemical paintings and engravings. Although he was no ornithologist, the birds in his garden were a source of much interest to him, and he had the gift of winning their confidence. The sight of a robin flying to his hand to take the morsel it had learnt to expect there revealed a side of his nature that was not generally known. He was musical and had played the violin in his youth, but he had no interest in games, and the only exercise he ever took was the walk between his house and the laboratory.

He bore the trials of his long illness with astonishing fortitude, and his indomitable will-power carried him through his duties until within a few months of his death.

W. H. MILLS

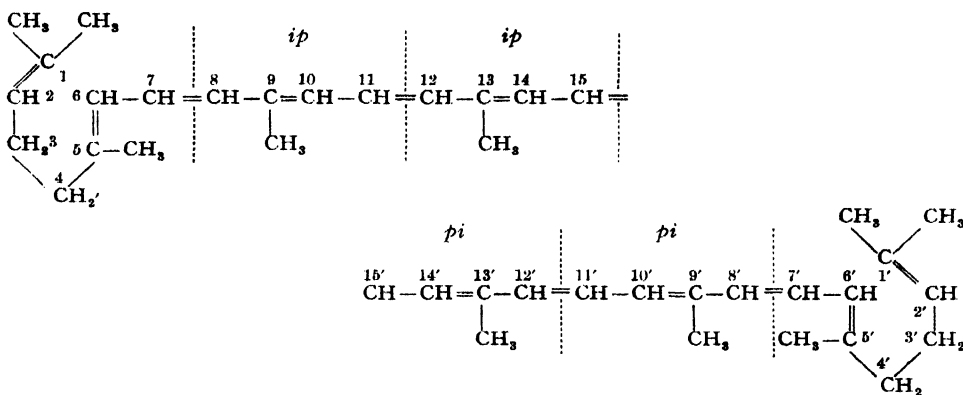
Carotene and Allied Substances in Foods and Feeding Stuffs

(Read at the Joint Meeting of the Society with the Food Group of the Society of Chemical Industry, February 7th, 1940)

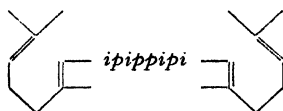
I. THE CONSTITUTION AND PHYSIOLOGICAL SIGNIFICANCE OF CAROTENE AND ALLIED PIGMENTS*

By R. A. MORTON, D.Sc., F.I.C.

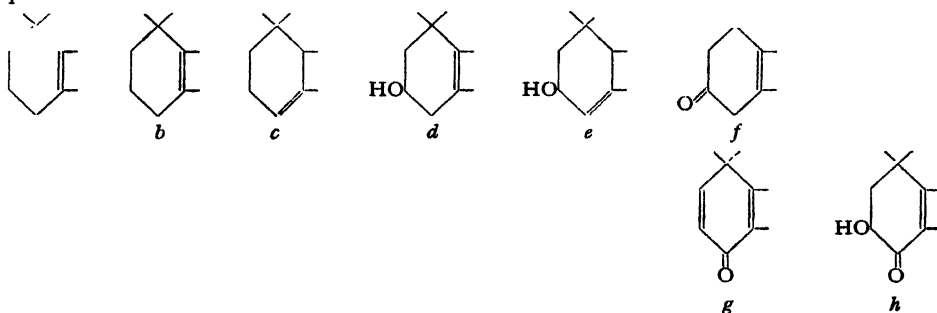
THE simplest carotenoid is lycopene, $C_{40}H_{56}$; on complete hydrogenation to perhydrolycopene, $C_{40}H_{82}$, thirteen bonds disappear. Quantitative degradation by means of ozone, permanganate and chromic acid as oxidising agents leads to the formula:—



The central polyene chain, $C_8-C_{8'}$ inclusive, is made up of four isoprene units arranged in pairs which are united in reverse order at C_{15} and $C_{15'}$, and may be abbreviated as *ipippi* (*ip* denoting an isoprene unit). Lycopene may thus be written:



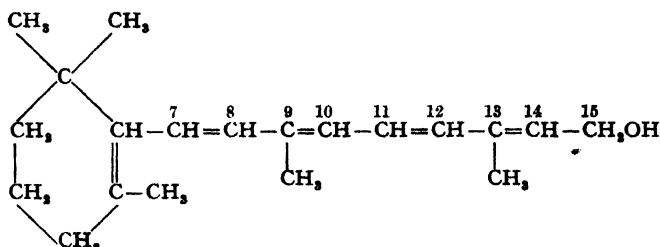
The polyene chain is common to a large number of carotenoids, but the terminal groups may consist of substituted or unsubstituted rings of the α - or β -ionone type as below:



* Condensed, by permission, from the summary in *Nature* (1940, 145, 286). The paper has been published in *Chemistry and Industry*, 1940, 59, 301-307.

Green leaves and other vegetable products contain α - and β -carotene and "xanthophyll," a mixture of hydroxylated carotenoids, zeaxanthin and lutein predominating. Their functions in plant physiology are not yet understood, but, owing to the connection between carotene and vitamin A, the position concerning the rôle of carotenoids in the nutrition of animals is much less obscure.

Vitamin A from fish liver oils ($C_{30}H_{48}OH$) possesses a constitution which differs from one half of the symmetrical β -carotene molecule only by the addition of the elements of the water:



The fact that green foodstuffs may cure avitaminosis A just as well as fish liver oils was explained when it was found that pure "carotene" undergoes fission *in vivo* with formation of vitamin A.

The only carotenoids which act as precursors or provitamins A are those that possess intact one half of the β -carotene molecule. They include those shown in the accompanying table, echinonene and a few derivatives prepared *in vitro* from natural provitamins. There is no evidence that animals can synthesise either provitamins A or vitamin A *de novo*, or that the conversion of carotene to vitamin A is reversible.

| Substance | Constitution | | Occurrence |
|----------------------|----------------------|----------|-------------------------------------|
| Lycopene | <i>a-iiippiipi-a</i> | | Ripe tomatoes. |
| * α -Carotene | <i>b</i> | <i>c</i> | Red palm oil, mountain ash berries. |
| * β -Carotene | <i>b</i> | <i>b</i> | Carrots, leaves, etc. |
| * γ -Carotene | <i>b</i> | <i>a</i> | Leaves of lily-of-the-valley. |
| * Kryptoxanthin | <i>b</i> | <i>d</i> | Yellow maize. |
| Zeaxanthin | <i>d</i> | <i>d</i> | Maize, egg yolk, leaves. |
| Lutein | <i>d</i> | <i>e</i> | Grass, green leaves. |
| * Myxoxanthin | <i>b</i> | <i>g</i> | } Algæ, especially blue-green algæ. |
| * Aphanin | <i>b</i> | <i>f</i> | |
| Rubixanthin | <i>d</i> | | |
| Astaxanthin | <i>h</i> | <i>h</i> | Crustacea. |

* Provitamins A.

The animal body contains only small quantities of carotenoids, and is not equipped to assimilate large doses. Carotene utilisation is optimal when minimal doses are fed in oil solution, and the transport of carotene through the intestinal wall is conditional on normal fat absorption. The site of the conversion of carotene to vitamin A is generally held to be the liver; certainly the liver is the main storage depot for the vitamin. In most species there is a normal level of concentration of vitamin A—and of carotenoid—in the blood. Carotenoids are also found in the pigmented layer of the eye and in yellow bone marrow.

Milk contains both vitamin A and carotenoids (largely β -carotene) and for a given species the total vitamin A activity of normal milk tends to be fairly constant, but in domesticated animals there are interesting variations with breed. Thus Holstein and Ayrshire cows yield milk with little carotene but more vitamin A,

whereas Guernseys give a cream more deeply coloured by carotene but less rich in vitamin A. The new-born possess very low liver reserves of vitamin A, and it is significant that colostrum may possess vitamin A activity one hundred times that of normal milk. Human colostrum is two or three times as potent as early milk, which in turn, is five or ten times as rich as the later milk.

The earliest sign of shortage of vitamin A or provitamin A is defective low-intensity vision. Visual purple, the photosensitive substance of the rods, may be obtained from retinas; it is a conjugated protein from which vitamin A can be separated. Faulty dark adaptation is due to delayed regeneration of visual purple, and in the majority of subjects can be remedied by supplementing the diet with vitamin A or carotene. In order to prevent night blindness in cattle, sheep, pigs, rats and horses, Guilbert finds that either some $25\text{--}30\mu\text{ g./kg.}$ body weight ($1\mu\text{ g.} = 10^{-6}\text{ g.}$) of β -carotene or $6\text{--}8\mu\text{ g./kg.}$ of vitamin A is needed daily.

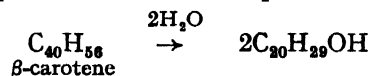
Vitamin A deficiency is characterised by widespread atrophy of epithelial structures and often by xerophthalmia, but retardation of growth (weight) is the criterion most readily amenable to quantitative interpretation.

The diet of most town dwellers shows inadequate vitamin A activity, especially during the winter months. It is also certain that most winter milk from stall-fed cattle is inferior to summer milk. Artificially dried grass is superior to hay both in respect of protein and provitamin A content, and its value as a feeding stuff has been established in many well-controlled experiments. The addition of vitamins A and D to margarine is a valuable way of alleviating vitamin deficiency, but the problem of utilising the available resources to the best advantage and of increasing the supply to meet the known needs has not been solved. That it is necessary and possible to do so cannot be doubted, nor that the cost of effective action would be a small fraction of the cost of inaction.

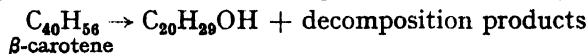
This raises the point of the relative efficiency of carotene and vitamin A. The accepted unit of vitamin A activity is that exerted by $0.6\mu\text{ g.}$ of pure β -carotene, so that the pure substance has a potency of $1.66 \times 10^6\text{ I.U./g.}$ (by definition). Vitamin A, according to the best available data, has a potency near $3.0\text{--}3.3 \times 10^6\text{ I.U./g.}$, whereas all provitamins other than β -carotene have an activity near $0.83 \times 10^6\text{ I.U./g.}$ These figures apply to rats receiving minimal doses.

There is urgent need for more research on the relative efficiencies of provitamins and vitamin A at the level needed to convert a marginal diet into an optimal diet.

The commonly accepted view that the equation



corresponds with a process occurring *in vivo* would lead to $1.56 \times 10^6\text{ I.U./g.}$ for pure vitamin A. This is not in accord with experience; it rests upon the assumption that fission occurs exclusively at C15—15' double bond, and its only advantage is that it agrees with the superiority of β -carotene over other provitamins. Unsymmetrical fission is more plausible, and an equation



is in closer harmony with the observed potency of vitamin A.

Vitamin A is estimated spectroscopically by utilising the absorption maximum at $325\text{m}\mu$. There is at present no reason to justify changing the accepted conversion factor, namely:

$$E_{1\%}^{1\text{cm}} 325\text{m}\mu, 1 = 1,600\text{ I.U./g.}$$

II. THE COMMERCIAL DETERMINATION OF CAROTENE AND ALLIED PIGMENTS, WITH ESPECIAL REFERENCE TO DRIED GRASS AND OTHER LEAFY MATERIALS

By W. M. SEABER, B.Sc., F.I.C.

THE work in our laboratory has been mainly concentrated upon the determination of carotene in dried grass, alfalfa and similar materials. In England, from a commercial point of view, α - and β -carotenes are the only substances of this class whose determination is usually required. In fact, it has been usual to include α -, β - and γ -carotenes as one substance under the name of "carotene." For most purposes this is convenient, and in grass and many leafy materials β -carotene is practically the only one present; the determination of total carotenoids is not often called for in commercial work. Xanthophyll is sometimes determined separately, but for approximate purposes can be taken as the difference between total carotenoids and carotene.

In this paper the main lines of the procedure adopted for the determination of the carotenes is outlined, and the process we have worked out in the laboratory of John Hughes is then described in detail. There are broad principles common to most of the classes of substances containing carotenes, and it is usually only the details of extraction that need be varied.

It was not until 1913 that serious attempts were made to distinguish between the various carotenoid pigments. In that year Monteverdi and Lubimenko reported a spectrographic method, and the well-known Willstätter and Stoll process was also put forward.¹ This consisted in extracting the tissue with acetone, saponifying the chlorophyll and removing it in the aqueous layer, and partitioning the xanthophyll and carotene between methanol and petroleum spirit. Most subsequent processes have been based upon Willstätter's procedure.

EXTRACTION OF PIGMENTS.—The processes can be divided into two groups: (a) those in which the pigments are extracted with a solvent and saponification is carried out subsequently, and (b) those in which the original substance is heated with alkali before extraction with a solvent.

In the first group numerous solvents have been proposed, including petroleum spirit, heptane, ethyl ether, acetone, methanol, ethanol (*e.g.* by Bolin and Khalapur,² and in the process of the Danish State Laboratory³), pyridine (Russell, Taylor and Chichester⁴; Smith and Smith,⁵) and *n*-butyl alcohol (proposed for flour by Binnington, Sibbitt and Geddes,⁶ who found that it had better extractive action than most of the other solvents investigated).

The use of mixed solvents (one of which is usually petroleum spirit) has advantages, especially if one of the solvents is removable by water. Examples are the use of acetone followed by ether (Schertz⁷), a mixture of 93 per cent. of light, cleaners' naphtha and 7 per cent. of ethanol (Ferrari⁸), benzine and methanol (Kuhn and Brockmann⁹), petroleum spirit and methanol (Campbell¹⁰), and our

own process, in which a mixture of 15 parts of acetone and 45 parts of petroleum spirit is used.

Grinding either with sand or in a small ball mill is often recommended. We have found grinding with sharp sand in a mortar satisfactory for dried grass, etc.

After extraction of the pigments it is usual to carry out a saponification process, followed by treatment with water. By this means, chlorophyllines and flavones are carried into the aqueous layer, and xanthophyll esters are split up, so that free xanthophylls are obtained, which can be removed from the petroleum spirit by the usual treatment with methanol. Kuhn and Brockmann⁹ apply the treatment with methanol before saponification. A very simple method of saponification is to shake the petroleum spirit solution at ordinary temperature with a 30 per cent. solution of potash in methanol, but sometimes it is preferred to heat under a reflux condenser, in order to make sure of splitting up the xanthophyll esters. So far as dried grasses, etc., are concerned, we can find no evidence that any xanthophyll esters remain undecomposed after the cold treatment. In any process in which acetone is used the acetone must be washed out before the treatment with hot alkali, otherwise brownish substances not removable by methanol are formed.

The main object of the processes in Group (b) is to break up tissue and allow the solvent to come into more intimate contact with the pigment. Coward¹¹ appears to have been the first to employ such a method. She ground with sand and heated with alcoholic or aqueous potash (the latter being preferred on account of less formation of resins) and then extracted with petroleum spirit. In our experience preliminary alkali treatment tends to give low results in some instances.

Probably the best known process in this class is that of Guilbert,¹² which was designed for forage. The sample is boiled for 30 minutes with a saturated solution of potassium hydroxide in ethyl alcohol, which is added in the proportion of 20 ml. per gram of the material, and then extracted with ethyl spirit. The ether is distilled, first at atmospheric pressure and finally under reduced pressure, and the residue is taken up in petroleum spirit for the subsequent methanol treatment.

It seems desirable to avoid the use of ethyl ether, not only to save time, but also because there is a risk of losing carotene. Petersen, Hughes and Freeman¹³ omit the ethyl ether and extract direct with petroleum ether (40–60° C.). Munsey, of the U.S. Food and Drug Administration, Washington, has published a collaborative investigation of the Petersen, Hughes and Freeman process, including various methods of measurement of the carotene in the final solution.¹⁴ Moon¹⁵ also discards the use of ether.

Some prefer to heat with aqueous potash, *e.g.* Coward,¹¹ Fergusson and Bishop,¹⁶ Moon.¹⁵ In all these processes, the main object is to get the yellow pigments finally into petroleum spirit, and remove the chlorophyll into the aqueous layer. The latter is drawn off (and, if necessary, re-extracted with petroleum spirit), and the petroleum spirit layer is washed a little and is then ready for the next step, which is usually a treatment with methanol containing a little water. The object of this process is to remove the so-called xanthophyll.

In the past there has been a little confusion as to the definition of xanthophyll, but it seems now to be accepted that there is a definite substance obtained from

leaves, eggs, and the petals of many flowers, to which the name xanthophyll was given by Karrer and Notthafft¹⁷; this compound, which was called lutein by Kuhn, Winterstein and Lederer,¹⁸ is a di-hydroxy derivative of α -carotene. The xanthophyll extract from leaves will consist mainly of that substance and zeaxanthin, but sometimes it may contain other derivatives of the so-called "phytoxanthins" of Karrer and Notthafft, and may include zeaxanthin, flavoxanthin, violaxanthin, taraxanthin, fucoxanthin, rhodoxanthin, capsanthin, crocetin, etc. From a commercial point of view, all these are included as xanthophylls (kryptoxanthin, which occurs in yellow corn, sunflower, capsicum, eggs, etc., is also classed as a xanthophyll, but is found mainly in the petroleum spirit layer).

Many workers (*e.g.* Schertz, Pyke¹⁹) used 85 per cent. methanol, but it seems now to be generally accepted that the best proportions are 10 vols. of water to 90 vols. of methanol, to give a strength of about 92 per cent. by volume (sp.gr. 0.830 at 15°/15° C.).

The petroleum spirit solution is shaken in a separating funnel with the methanol, which is drawn off, and the process is repeated until the last washing is practically colourless. If desired, a colour reading of the petroleum spirit solution can be made before the methanol treatment; this gives a result which is usually called "total carotenoids," the value generally being calculated in terms of β -carotene. This is a partition process, and presumably a small quantity of carotene goes into the methanol layer, and the xanthophylls to a small extent into the petroleum spirit layer. Miller²⁰ found 6 per cent. of the xanthophyll in the petroleum spirit layer, when using 92 per cent. methanol (by volume). According to our experiments, the quantity of carotene dissolved in methanol of sp.gr. 0.830 under these conditions is very small indeed.

The tendency is for the hydrocarbons (*e.g.* carotene, lycopene) and the less highly oxygenated xanthophylls (*e.g.* kryptoxanthin) to stay mainly in the petroleum spirit phase, while the more highly oxygenated xanthophylls go into the methanol phase (note, however, that some rhodoxanthin may go into the petroleum spirit).

In the ordinary commercial processes the coloured substances in the petroleum spirit layer are counted as carotene, and, if this is all that is required, it is only necessary to make up to a suitable volume and determine the colour value in terms of β -carotene, or use the spectrograph at an agreed wave-length.

The solution thus obtained is not pure carotene, however, and in a strict analysis it is necessary to go further. Sometimes spectrophotometric methods may be used, but often further separation is possible only by chromatographic methods. A few notes on spectroscopic methods may be of interest, although the equipment is not usually found in the average commercial laboratory. Many chemists have worked out methods depending upon absorption spectroscopy, both with visual and ultra-violet spectroscopes. (Gillam, Heilbron, Morton, Bishop and Drummond²¹ did a large amount of work in this direction, using chloroform solutions, and taking readings before and after the addition of an antimony trichloride reagent.) Usually the methanol separation is first carried out, and the spectroscope is used to examine the separate portions, but Miller^{22, 23, 24, 25, 26}

has worked out methods for two, three and even four constituents in presence of each other, and by using his methods it might be possible with some materials to work on solutions that have had no preliminary methanol separation. Miller gives examples of the use of the method for determining mixtures of α - and β -carotenes.

Wave-length $450m\mu$ is suitable for the determination of β -carotene. In our laboratory we have obtained E values for two separate supplies of β -carotene. One of these was kindly sent by Professor A. C. Chibnall. Our value at $450m\mu$ for $E_{1\text{cm.}}^{1\%}$ was 2520 in petroleum spirit. This is very close to that of Gillam,²⁷ who obtained 2500 in petroleum spirit and 2200 in chloroform, both at $450m\mu$. Gillam in the same paper gives maxima for various carotenoids.

Working with a sample of S.M.A. β -carotene from Cleveland, Ohio, we obtained $E_{1\text{cm.}}^{1\%}$ 2440 (petroleum spirit), which agrees very well with the values generally given in the United States.

Petersen, Hughes and Freeman¹⁸ employ three wave-lengths, viz. 4550, 4700 and 4800\AA and calculate the carotene from the average.

Among recent workers employing spectrography may be mentioned Clark and Gring,²⁸ who determined β -carotene and kryptoxanthin together in yellow corn. Shrewsbury, Kraybill and Withrow²⁹ describe the use of a photo-electric photometer and of a Bausch and Lomb spectrophotometer.

CHROMATOGRAPHIC METHODS.—Chromatographic methods are rather difficult to deal with in any systematic way, as they depend upon somewhat obscure adsorptive properties of inorganic materials. Twsett was one of the earliest workers with this method. For full details, reference should be made to such works as those of Zechmeister and Cholnoky,³⁰ and of Winterstein,³¹ and to articles by Wiseman and Kane, Strain and others in the *Journal of Biological Chemistry* and the *Biochemical Journal*. Briefly, the method consists in dissolving the mixed pigments in a suitable solvent (frequently petroleum spirit), and passing the solution through a well-packed column of a suitable adsorbent. Usually the pigments will distribute themselves in bands according to their adsorption affinities.

Frequently the bands can be made to separate by washing with the solvent, or by pouring a mixed solvent through the tube (for example, a mixture of petroleum spirit and benzene). The bands can then be dealt with by pushing the column up the tube and removing them as they appear at the top, or, with narrow tubes, the tube can be cut off at suitable places. In a very elaborate analysis, the various substances can then be extracted from the adsorbent, and their absorption curves by means of the spectroscope.

The most usual adsorbents are alumina (see Willstaedt and With³²), lime, calcium hydroxide, calcium carbonate and magnesia. As a general rule, the most oxygenated carotenoids are the most strongly adsorbed, and will be found in the top of the tube, while the pigments with less oxygen and the hydrocarbons, such as lycopene and α - and β -carotenes, will tend to pass to the bottom of the tube. As α -carotene is more loosely held than β -carotene it can often be washed out of the tube with solvents. In obtaining the separations it is sometimes an advantage to mix active and inactive adsorbents, e.g. the use of 1 part of Merck's alumina

with 1 or 2 parts of ordinary anhydrous alumina allows α -carotene to be washed down more readily. Frequently it is difficult to get more than roughly quantitative results by this method, but we have worked out a modification which we have found very useful in the examination of dried grass. This is described later.

The following are some examples of the use of this method in the separation of carotenoids. (1) Buxton⁸³ separated kryptoxanthin from carotene by dissolving the pigments in heptane, and using calcium carbonate activated at 200° to 300° C. (2) Hegsted, Porter and Petersen⁸⁴ removed non-carotene pigments from silage by passing the solution in "Skellysolve" through calcium carbonate. (3) Violaxanthin is separated from lutein and zeaxanthin by means of calcium carbonate (Kuhn and Brockmann⁹). (4) Lycopene is separated from β -carotene by dissolving in benzene, and running the solution through a mixture of activated magnesia and Supercel (Strain). (5) Coward¹¹ passed a petroleum spirit solution from tomatoes through calcium carbonate, and found that carotene washed through first, then lycopene. (6) Lime, or calcium hydroxide, has often been used to separate α - from β -carotene; the latter is said to form a top red layer, and the former a lower yellow layer. Strain separated α - and β -carotenes by the use of a mixture of 1 part of activated magnesia and 1 part of Supercel. (7) Zechmeister⁹⁰ separated kryptoxanthin, lycopene, γ -carotene, β -carotene and α -carotene, in that order, with alumina.

My own experience with adsorbents other than Merck's alumina has been disappointing. We have found filtration very slow, and adsorption very much inferior to that obtained with the alumina. Our experiments with alumina are described later, but I will give here the process worked out in our laboratory for the routine determination of carotene in dried grasses.

ROUTINE DETERMINATION OF CRUDE CAROTENE.—From 0.25 to 0.5 g., according to the richness of the sample, is thoroughly ground with about 20 times its weight of sharp silver sand (60-mesh sieve) in a suitable mortar until a soft powder is obtained. This is transferred to an extraction thimble lined with filter-paper, the mortar being rubbed out with a little sand which is added to the ground powder. A plug of cotton-wool is placed in the top of the thimble and the sample is extracted in a continuous-drip type of extractor (this is preferable to the Soxhlet type) with a mixture of 15 ml. of acetone and 45 ml. of petroleum spirit for at least one hour, or until no more colour is extracted. The residue can be re-ground and re-extracted. The extract is cooled and transferred to a separating funnel, and the extraction flask is rinsed with first 3 ml. and then 2 ml. of petroleum spirit. Five ml. of a 30 per cent. solution of potassium hydroxide in pure methyl alcohol (this solution must be quite colourless) are added, and the funnel is shaken very thoroughly for at least two minutes, after which 200 ml. of water are added and the funnel is very gently inverted once or twice. The aqueous layer is run off, and the petroleum spirit layer is shaken vigorously with 200 ml. of water. In this way chlorophyll and flavones are removed.

Xanthophylls are removed from the petroleum spirit solution by shaking successively with 30, 15 and 15 ml. of methyl alcohol of sp.gr. 0.830 at 15°/15° C. (made by mixing 90 volumes of absolute methyl alcohol with 10 volumes of water

separately measured). The petroleum spirit solution is made up to 50 ml. or to such other volume as is convenient for colour measurement. To ensure brightness, about 1 g. of anhydrous sodium sulphate may be added.

There is nothing especially novel about this process, but our aim has been to arrange the manipulation so that it is workable for a large number of routine samples. With slight modification, it has been accepted tentatively as the official process of the Grass Driers' Association, as a result of the investigations of a Committee of Chemists called together by Mr. Davies, a member of the Council of that Association; the report of their findings will be issued very shortly.

In our process, we have followed Pyke¹⁹ by working with small quantities. Extraction is easy, and the whole manipulation is much simpler than when larger quantities are taken. As regards the solvent, there is an advantage in using acetone as part of the mixture, on account of its low b.p. The residue is usually colourless after an hour's extraction, but, if any doubt exists, it is easy to re-grind and continue the extraction for a while.

We have also tried a variation of the process by shaking from 0.5 to 1 g. of the grass (after grinding with sand) with 75 ml. of petroleum spirit and 25 ml. of acetone for 1 hour on a mechanical shaker. An aliquot part is taken, and the remainder of the process is as described. Extraction appears to be complete under these conditions, and the procedure has the advantage that the whole operation is carried out at the ordinary temperature.

There seems to be no need to re-extract the aqueous portion. Whenever this has been tried, the second extract has invariably been practically colourless. It may be desirable with very rich grasses to make a second treatment with 30 per cent. potassium hydroxide solution, but residual traces of chlorophyll do not seem to interfere with the determination of the carotene, provided that the first shaking has been very thorough.

COLORIMETRIC ESTIMATION.—It is very difficult, in England at any rate, to get pure β -carotene for use as a standard, and, if obtainable, it is very expensive; moreover, it is practically impossible to keep a solution without change. On this account it has been proposed to use as standards more stable solutions, which can be compared with pure carotene solutions, and the values so obtained used for all subsequent determinations.

Azobenzene has been used by Kuhn and Brockmann. A solution of 14.5 mg. in 100 ml. of 95 per cent. alcohol was said to be equivalent to one containing 0.235 mg. carotene per 100 ml. in light petroleum. Potassium dichromate is the substance most commonly employed, and has been used in various strengths by different workers. Fraps used a 0.1 per cent. solution, and Russell a 0.036 per cent. solution. Ferguson³⁶ used the Klett colorimeter, with 0.1 per cent. solution. Under those conditions his figure for a 0.025 per cent. solution works out at 0.150 mg. of carotene per 100 ml.

Lovibond Readings.—Ferguson has also constructed a curve for Lovibond readings plotted against carotene. His reading for a solution of carotene at 0.1 mg. per 100 ml. is 1.6 yellow units.

The Committee of the Grass Driers' Association, referred to above, used a 0.025 per cent. solution of potassium dichromate, and agreed upon the figure of

0.158 mg. of carotene per 100 ml. of petroleum spirit as the equivalent carotene solution. (This agrees well with the figure 0.160 given by Connor.³⁶) This comparison was obtained by means of the Lovibond Tintometer, but with glasses that were probably of very different ages. During the course of the investigations, it was found that individual observers obtained widely different results in reading, particularly in the higher ranges. Even with a carotene solution containing 0.1 mg. per 100 ml. values ranging from 1.6 to 2.2 were obtained, with an average of about 2.0. It was found, however, that if observers constructed their own curves, using either a carotene solution or potassium dichromate, much better agreement could be obtained on the actual carotene figure for any sample. In using the Lovibond instrument it is usual to match with yellow and red, and to count only the yellow reading. The red reading is usually very small, but most operators find that it helps them to get a closer match.

We always prefer to read at a point not higher than about 3.0 yellow Lovibond. With very careful work it is possible to get good results with the Lovibond instrument, but we much prefer a photoelectric instrument.

Photoelectric Comparison.—Actually, most of our work has been carried out by means of the Spekker photoelectric absorptiometer. It appears to be generally accepted that the graph showing the relation between carotene and potassium dichromate by various colorimeters is a straight line, provided that the concentrations are not too great, and we have found that this applies to the Spekker instrument. With that instrument we obtained a straight line by plotting dichromate strengths against drum readings, but the relation of carotene to dichromate was a little higher than the figure given above for Lovibond, *viz.* a 0.025 per cent. solution of potassium dichromate was equivalent to a carotene solution containing 0.165 mg. per 100 ml.

TABLE I

| Crude "Carotene" (calculated on dry sample) p.p.m. | | | | Crude "Carotene" (calculated on dry sample) p.p.m. | | | |
|--|----|----|-----|--|----|----|-----|
| Carrot | .. | .. | 720 | Barley shoots | .. | .. | 155 |
| Dried carrot meal | .. | .. | 500 | Spinach | .. | .. | 600 |
| Wheat flour (bleached) | .. | .. | 1 | Turnip leaves | .. | .. | 550 |
| Silage | .. | .. | 140 | " stalks | .. | .. | 40 |
| " | .. | .. | 230 | Tomato (green) | .. | .. | 55 |
| " | .. | .. | 200 | " (pink) | .. | .. | 110 |
| " (red clover) | .. | .. | 209 | " (red) | .. | .. | 220 |
| " (white clover) | .. | .. | 523 | Hay | .. | .. | 90 |
| " | .. | .. | 378 | Palm oil | .. | .. | 920 |

Our working plan has been to set our instrument by means of β -carotene and to keep a stock (0.025 per cent.) solution of potassium dichromate, which is read from time to time, in order to ensure that no change has taken place.

Other special instruments in common use are the Bolton-Williams Absorptiometer, and the Zeiss-Pulfrich visual absorptiometer; also various types of visual colorimeters, such as the Klett and the Duboscq instruments mentioned

above. Direct comparison in Nessler tubes can, of course, be used, but in our experience it is difficult to get consistent results by this method.

In Table I, some examples of our determination of crude carotene in different substances are given.

TABLE II
EXAMPLES OF TREATMENT BY SPUR'S CHROMATOGRAPHIC METHOD FOR DRIED
GRASS AND ALFALFA

| Sample | | | "Carotene" by the John Hughes method Before treatment | | Carotene After treatment | Ratio |
|-------------------------------|----|----|---|--|--------------------------------|-------|
| | | | | | | |
| Grass meal, 1185 | .. | .. | 190 | | 135 | 0.70 |
| " " 1186 | .. | .. | 425 | | 315 | 0.74 |
| " " 1187 | .. | .. | 400 | | 325 | 0.81 |
| " " 1188 | .. | .. | 275 | | 220 | 0.80 |
| " " 1189 | .. | .. | 180 | | 130 | 0.72 |
| " " 1190 | .. | .. | 240 | | 175 | 0.73 |
| " " 1191 | .. | .. | 250 | | 195 | 0.78 |
| " " 1192 | .. | .. | 320 | | 225 | 0.70 |
| " " 1193 | .. | .. | 160 | | 125 | 0.78 |
| " " 1194 | .. | .. | 175 | | 140 | 0.80 |
| " " 1195 | .. | .. | 300 | | 235 | 0.78 |
| " " 1196 | .. | .. | 390 | | 275 | 0.71 |
| " " 1199 | .. | .. | 255 | | 200 | 0.78 |
| " " 1200 | .. | .. | 245 | | 200 | 0.82 |
| Grass meal and molasses, 1198 | | | 145 | | 115 | 0.79 |
| " " " " 1203 | | | 400 | | 310 | 0.77 |
| Grass meal, 1210 | .. | .. | 250 | | 220 | 0.88 |
| " " 1211 | .. | .. | 270 | | 220 | 0.82 |
| " " 1218 | .. | .. | 535 | | 410 | 0.76 |
| Lucerne leaf, 1207 | .. | .. | 310 | | 275 | 0.89 |
| " stalk, 1208 | .. | .. | 205 | | 193 | 0.94 |
| " meal, 1230 | .. | .. | 220 | | 190 | 0.86 |
| " " 1243 | .. | .. | 430 | | 320 | 0.75 |
| Canadian alfalfa, A.663 | .. | .. | 175 | | 160 | 0.91 |
| " " A.664 | .. | .. | 165 | | 145 | 0.88 |
| Silage, 297 | .. | .. | 92 | | 50 | 0.54 |
| " (red clover), 250 | .. | .. | 65 | | 45 | 0.70 |
| " (white clover), 251 | .. | .. | 130 | | 102 | 0.78 |

Chromatographic Procedure.—Reference has already been made to the chromatographic procedure we have adopted in arriving at some estimate of the purity of the final solutions obtained in our process. Our first investigations along this line were due to Mr. Bernard Spur of Copenhagen, who showed us this method of using Merck's alumina. In his method, the petroleum spirit solution after the usual alkali and methanol treatments is washed free from methanol and drawn through a column of Merck's alumina in a tube about 2 cm. in diameter; β -carotene is adsorbed to form a red layer, and above it can be seen a yellow layer. The adsorbed layers are developed with a mixture of 1 part of petroleum spirit and 1 part of benzene, and then the whole column is pushed upwards, and the yellow layer is removed as it comes up.

The red layer of β -carotene is dissolved from the alumina by means of a solution of 10 per cent. ethanol in petroleum spirit. The solution is made up to definite volume, and the carotene is determined by the usual methods. In Spur's method, we prefer to use a very small tube, about 4 mm. in diameter, and about 5 or 6 cm. long. This serves for the usual quantities obtained from 0.25 or 0.5 g. of dried grass. After passage of the solution and development of the bands, the tube is cut at a point in the yellow band just above the red, and the yellow is scraped off, until it is all removed and the red appears. There is always a little doubt as to the sharpness of this mechanical separation; also, there seems a tendency for the outside portion of the red to change to yellow.

In experiments to discover a more convenient method we found that the addition of 3 per cent. of acetone to the solution before passing through the alumina caused the carotenes to be carried through, while the yellow substance remained in the alumina. The tube can be washed with a small quantity of 3 per cent. solution of acetone in petroleum spirit, and the solution of carotenes that comes through can be dealt with in the usual way.

Table II shows some results obtained by Spur's method, and Table III gives comparative results obtained by Spur's method and by the 3 per cent. acetone method. These agree well as a rule, and it is probable that the yellow bands obtained by the two methods are identical.

It is difficult to ascertain the nature of the yellow bands. In one test an ultra-violet spectrograph was taken, but I was unable to identify them. In any event the substance is definitely non-carotene pigment. With grasses, the unadsorbed portion appears to be homogeneous, as judged by the results of washing it and passing it through a tube packed with one part of Merck's alumina and one part of ordinary alumina, and it is reasonable to assume that it is β -carotene.

TABLE III

EXAMPLES OF THE THREE PER CENT. ACETONE PROCESS APPLIED TO THE
"CAROTENE" SOLUTION OBTAINED BY THE JOHN HUGHES METHOD

| | Original | 3 per cent. acetone process | Modified Spur's method |
|----|----------|--------------------------------|---------------------------|
| 1. | 470 | 385 | 370 |
| 2. | 385 | 350 (repd. 350) | — |
| 3. | 380 | 320 | 300 |
| 4. | 240 | 200 | 210 |
| 5. | 280 | 190 | 180 (old sample) |
| 6. | 250 | 170 | 190 (old sample) |

Comparison between 3 per cent. acetone treatment:

(a) on original green solution,

(b) on yellow solution after removal of chlorophyll and xanthophyll.

| | (a) p.p.m. | (b) p.p.m. | |
|----|--------------------|---------------|------------|
| 1. | 190 (repeated 180) | 190 | (Spur 180) |
| 2. | 320 | 300 | |
| 3. | 325 | 305 | |

Note.—All the above figures represent milligrams per kilogram.

It will be seen that the ratio between this pure carotene and the crude carotene varies from 0.7 to 0.8, though apparently for old grasses the proportion may fall still lower than 0.7.

To explore the possibilities of simplification, we applied the 3 per cent. acetone process at an earlier stage, *i.e.* on the original extract of the grass. We washed out the acetone by means of a sodium sulphate solution (made by adding 1 part of saturated sodium sulphate solution to 1 part of water) with gentle shaking, as emulsions sometimes form. We then added 3 per cent. of acetone to the petroleum spirit solution, and passed it through the alumina (a rather longer column than usual is safer, say 7 cm.) and washed with a little of the 3 per cent. solution of acetone in petroleum spirit. The amount of carotene obtained by this method agrees very well with that obtained by passing the solution, after treatment with alkali and methanol, through the alumina, as all the xanthophyll and chlorophyll are removed, as well as the non-carotene substances already mentioned. Occasionally, a trace of chlorophyll may escape, but it can be held back by passing the solution through a fresh tube of alumina.

As a further simplification, 0.5 g. of the grass was added to 100 ml. of 3 per cent. acetone solution and shaken on a mechanical shaker for 2 hours. An aliquot portion (25 ml.) was taken, and passed through the alumina. The carotene-content thus found was practically identical with that obtained by making the usual extraction with a 25 per cent. solution of acetone in petroleum spirit, washing away the acetone, adjusting the solution to contain 3 per cent. of acetone, and passing it through the alumina as usual (Table IV).

TABLE IV

EXAMPLES OF REPETITIONS BY THE THREE PER CENT. ACETONE PROCESS

- (1) 0.5 g. of grass meal shaken with 100 ml. of a 3 per cent. solution of acetone in petroleum spirit for 2 hours on a mechanical shaker at ordinary temperature, and aliquot parts of 25 ml. each passed through alumina.

| Length of tube cm. | Readings on Spekker instrument | Carotene mg. per kg. |
|-----------------------|-----------------------------------|-------------------------|
| 5 | 0.15, 0.16, 0.16 | 330— |
| 7.5 | 0.16, 0.16 | 330 |
| 10 | 0.165, 0.16 | 330+ |

- (2) Shaken with 3 per cent. of acetone in petroleum spirit during 2 hours at 40° C.

| Length of tube cm. | Readings on Spekker instrument | Carotene mg. per kg. |
|-----------------------|-----------------------------------|-------------------------|
| 7.5 | (mean) 0.16 | 330 |
| 10 | (mean) 0.165 | 340 |

- (3) Extracted with 25 per cent. of acetone in petroleum spirit in a drip extractor; acetone washed out and petroleum spirit solution made to 3 per cent. acetone strength.

| Length of tube cm. | Readings on Spekker instrument | Carotene mg. per kg. |
|-----------------------|-----------------------------------|-------------------------|
| 7.5 | (mean) 0.17 | 350 |

More work is necessary to find out whether this extremely simple process could be generally applied to dried grasses. There would be the difficulty, of course, at the present time, that Merck's alumina is unobtainable, but no doubt an activated alumina of suitable quality could be prepared, or some other adsorbent used.

In this connection reference may be made to an interesting paper recently published by Fraps, Kemmerer and Greenberg³⁷ upon the adsorbent properties of magnesia and magnesium carbonate. It is very difficult to predict what any particular batch of magnesia is going to do, but the authors give some hints upon control. Since reading that paper, I have used a sample of light magnesia in connection with my 3 per cent. acetone process. Although filtration was much slower than with Merck's alumina, it was workable, and the results appear to be practically identical.* Further work along these lines should be useful.

PROCEDURE FOR SPECIAL MATERIALS.—The determination of carotene in dried grass, alfalfa and similar materials has been fully treated above. As regards wet grass, our process can be applied with the modification that plain acetone is first used to extract the sample to remove the water, after which the material is ground with sand and extracted with petroleum spirit.

We have examined spinach and several other leafy materials by our process; it also seems to work well with carrots and tomatoes. The 3 per cent. acetone process appears to allow β -carotene to pass through the alumina tube, while the lycopene (the chief pigment of tomatoes) is held as a purple-red layer, and can be dissolved out with a mixture of petroleum spirit and alcohol, and estimated by its colour value or by the spectrograph.

Palm Oil.—In this and similar fatty substances, extraction coincides obviously with saponification. Our cold saponification process worked satisfactorily for this, using 30 per cent. methanolic potash at the rate of 15 ml. for each gram of oil, then 50 ml. of petroleum spirit, diluting with water, removing the aqueous layer, and repeating the treatment with 7.5 ml. of potash solution. In the samples we have examined we have found no pigments other than α -carotene and β -carotene.

Butter.—A very simple approximate process has been put forward by Baumann and Steenbock.³⁸ They separate the fat and determine the extinction coefficients at $485m\mu$ and $460m\mu$ at 30°C . These are compared with the coefficients of β -carotene in refined cottonseed oil at 30°C .

A saponification process is described by Morton.³⁹ If desired, the xanthophyll can be removed by methanol as usual, but it is frequently assumed that about 6 per cent. is present, and a suitable deduction made. The reading of the final petroleum spirit solution can be made colorimetrically or spectrophotometrically. Many papers on this subject have been published by Gillam, Morton, Heilbron, Drummond and others in the *Biochemical Journal*.⁴⁰

We have applied successfully cold saponification with a 30 per cent. solution of potash in methanol. When using the process with 3 per cent. solution of acetone

* Since writing the above I have tried a sample of heavy magnesia kindly sent me by Mr. D. J. Campbell. This gave a better filtration than the light magnesia, but it was necessary to use a 7 per cent. solution of acetone in petroleum spirit.

It appears that the percentage of acetone in the petroleum spirit must always be adjusted to suit the adsorbent.

in petroleum spirit, only a very small yellow layer was found, and the pigment appeared to be practically all α - and β -carotenes; Gillam and Heilbron,⁴¹ however, found small quantities of kryptoxanthin.

Egg Yolk.—We have not done much work with this substance, but in one experiment we mixed the sample with sand and extracted it first with acetone. The residue was taken from the cartridge, ground and then extracted with a mixture of petroleum spirit and acetone. Hot saponification appeared to be necessary. In one experiment, working with the 3 per cent. acetone process, we found 0.7 p.p.m. of carotene in the yolk of an egg, and approximately the same proportion of kryptoxanthin.

Flour.—Kent-Jones used petrol for extracting the carotenoids. Ferrari used a mixture of 7 per cent. of ethyl alcohol with light, cleaner's naphtha (93 to 160° C.), whilst Binnington, Sibbett and Geddes⁶ used *n*-butyl alcohol (saturated with water to avoid turbidity), and claimed that better extraction was obtained than with the usual solvents.

Apparently it has generally been believed that nearly all the yellow pigment in flour is carotene, but Munsey⁴² claims that, although the total pigment may amount to from 2 to 3 p.p.m., the carotene is not more than 0.2 p.p.m. He uses a hot saponification process on the lines of that of Petersen and Hughes, and believes that the main portion of the pigment is xanthophyll or something very similar, and that previous high results for carotene were obtained because xanthophyll esters were not properly saponified and were found in the petroleum spirit layer.

Experiments on a few flours with our 3 per cent. acetone process confirm Munsey's statement, that the amount of β -carotene is very small (probably of the order of 0.15 per cent.) but in the particular flours examined I found very little xanthophyll after either hot or cold saponification. In one experiment the evidence seemed to point to the presence of lycopene, and in another to kryptoxanthin.

Yellow Corn.—Considerable attention has been given in the United States to the carotene content of yellow corn. This has been referred to above. Kryptoxanthin forms the major portion of the petroleum spirit phase in the partitioning with methanol. Our process appears to work well for this substance, and the 3 per cent. acetone treatment seems to separate the kryptoxanthin from the carotene.

CONCLUSIONS.—A process such as is used in our ordinary routine gives a useful commercial valuation, in spite of the fact that it does not determine the actual carotene and that the true carotene is sometimes only 80 per cent. of the figure obtained. There is some evidence that this percentage sinks with the age of the stored grass, and this suggests that the yellow substance may be an oxidation product of carotene.

I have shown that the use of Merck's alumina in the 3 per cent. acetone process appears to be a very promising method, capable of giving in an extremely simple way the true carotene percentage in dried grass and perhaps in many other materials. The great disadvantage is that Merck's alumina is now unobtainable;

possibly, however, magnesia, some experiments with which I have mentioned, may eventually take its place.

I wish to thank our staff for their assistance, and in particular Mr. Ward, who worked out most of the details of the process, and Mr. Rabnott for spectrographic and general research work.

REFERENCES

1. R. Willstätter and A. Stoll, "*Untersuchungen über Chlorophyll*," Springer, Berlin, 1913.
2. D. Bolin and A. M. Khalapur, *Ind. Eng. Chem., Anal. Ed.*, 1938, **10**, 417.
3. Private communication.
4. W. T. Russell, M. W. Taylor and D. F. Chichester, *Plant Physiol.*, 1935, **10**, 325.
5. L. L. W. Smith and O. Smith, *Plant Physiol.*, 1931, **6**, 265.
6. D. S. Binnington, L. D. Sibbitt and W. F. Geddes, *Cereal Chemistry*, 1938, **15**, 119.
7. F. M. Schertz, *Plant Physiol.*, 1928, **3**, 211.
8. C. G. Ferrari and C. H. Bailey, *Cereal Chemistry*, 1933, **10**, 277.
9. R. Kuhn and H. Brockmann, *Z. physiol. Chem.*, 1932, **206**, 41.
10. D. J. Campbell, Private communication.
11. K. H. Coward, *Biochem. J.*, 1924, **18**, 1114.
12. H. R. Guilbert, *Ind. Eng. Chem., Anal. Ed.*, 1934, **6**, 453.
13. W. J. Petersen, J. S. Hughes and H. F. Freeman, *Ind. Eng. Chem., Anal. Ed.*, 1937, **9**, 71.
14. V. E. Munsey, *J. Assoc. Off. Agric. Chem.*, 1939, **22**, 664.
15. F. E. Moon, *J. Agric. Sci.*, 1939, **29**, 295.
16. W. S. Ferguson and G. Bishop, *ANALYST*, 1936, **61**, 515.
17. P. Karrer and A. Notthafft, *Helv. Chim. Acta*, 1931, **15**, 1195.
18. R. Kuhn, A. Winterstein and E. Lederer, *Z. physiol. Chem.*, 1931, **197**, 141.
19. M. Pyke, *J. Soc. Chem. Ind.*, 1936, **55**, 1397.
20. E. S. Miller, *J. Amer. Chem. Soc.*, 1935, **57**, 347.
21. A. E. Gillam, I. M. Heilbron *et al.*, *Biochem. J.*, 1933, **27**, 878.
22. E. S. Miller, *Cereal Chemistry*, 1938, **15**, 310.
23. ——— *Plant Physiol.*, 1934, **9**, 681.
24. ——— *J. Amer. Chem. Soc.*, 1935, **57**, 347.
25. ——— *Bot. Gazette*, **96**, 447.
26. ——— *Plant Physiol.*, 1937, **12**, 667.
27. A. E. Gillam, *Biochem. J.*, 1935, **29**, 1831.
28. G. L. Clark and J. L. Gring, *Ind. Eng. Chem., Anal. Ed.*, 1937, **9**, 271.
29. C. L. Shrewsbury, H. R. Kraybill and R. B. Withron, *Ind. Eng. Chem., Anal. Ed.*, 1938, **10**, 253.
30. L. Zechmeister and L. V. Cholnoky, "*Die Chromatographische Adsorptionsmethode*," Springer, Vienna, 1937.
31. A. Winterstein, "*Prinzepts Chromatographische Adsorptionanalyse*."
32. H. Willstaedt and T. K. With, *Z. physiol. Chem.*, 1938, **253**, 40.
33. L. O. Buxton, *Ind. Eng. Chem., Anal. Ed.*, 1939, **11**, 128.
34. D. M. Hegsted, J. W. Porter and W. H. Petersen, *Ind. Eng. Chem., Anal. Ed.*, 1939, **11**, 256.
35. W. S. Ferguson, *ANALYST*, 1935, **60**, 680.
36. C. L. Connor, *J. Biol. Chem.*, 1928, **77**, 619.
37. G. S. Fraps, A. R. Kemmerer and S. M. Greenberg, *Ind. Eng. Chem., Anal. Ed.*, 1940, **12**, 16.
38. C. A. Baumann and H. Steenbock, *J. Biol. Chem.*, 1933, **101**, 547.
39. R. A. Morton, "*The Application of Absorption Spectra to the Study of Vitamins and Hormones*."
40. *E.g.* that by Gillam, *Biochem. J.*, 1934, **28**, 79.
41. A. E. Gillam and I. M. Heilbron, *Biochem. J.*, 1935, **29**, 835.
42. V. E. Munsey, *J. Assoc. Off. Agric. Chem.*, 1938, **21**, 331.
43. Neish, McFarland and Brechin, *Canadian J. Research*, 1939, **17**, [10], 325.

March 9th, 1940

DISCUSSION

Mr. E. B. ANDERSON said that, as Chairman of the Food Group, he would like to convey to the Society the appreciation of the Group for this joint meeting, which had now become an annual event. His knowledge of carotene was concerned with its connection with the colour of the cream of milk. It was well known that

in the winter, owing to the lack of colour in the cream, there were frequent complaints from customers that there was no cream on the milk. A year or two ago experiments were made on the use of dried grass for feeding cattle in winter, to see if a milk-cream with the colour of summer milk could be obtained. This feeding, however, had no effect whatever on the colour. They had found that the colour dropped until about the beginning of December and then remained constant until the cows were put out to grass, when an immediate improvement was shown. In the experiments, one herd was given the ordinary winter feed and two others dried grass, and the results were the same for all three herds. It was well known to most users of the Lovibond Tintometer that the inaccuracies were greatest when measuring colours in the yellow region.

Dr. S. K. KON said that, by a coincidence, he had with him a copy of a paper, prepared by himself and Mr. Thompson, which contained exactly the same findings as those of Mr. Seaber, and he could confirm those results in every detail. They had extracted the pigment and had had it analysed for them by Dr. Gillam, and now Dr. Morton was of opinion that it was an oxidation product of carotene. They were carrying out biological tests to ascertain if it was biologically active. With regard to the feeding of dried grass to cattle, they had carried out some experiments at Shinfield with dried grass, kale and mangolds in the quantities:—dried grass 14 lbs., kale 46 lbs., and mangolds 42 lbs. There was a very marked difference in the results; the milk from the cows fed with the dried grass contained twice as much carotene as the milk from the cows fed on kale and some five times more than the milk of the mangold cows. Of course, this was only an experiment on a small scale.

Mr. H. PRITCHARD said that Mr. Seaber had mentioned the difficulty of obtaining Merck's alumina; this question had also been raised at a meeting in the North. He would suggest that old supplies of alumina should be conserved as much as possible, and to this end his practice was to retain the alumina which had been used for chromatography and burn off all organic matter in a muffle furnace at 450° to 600° C. By this means the alumina was rendered quite white, its activity was unimpaired, and it could be used again many times. He had tried other aluminas. One supplied by a British firm seemed to contain a certain amount of water in combination, which made the rate of percolation of the solvent through the column very slow.

Dr. R. G. DAVIES, referring to Mr. Anderson's remarks on the feeding of grass to cows, said that he thought that results depended largely on the breed of cow. With Holstein cows the colour of the milk was sometimes changed, whereas with Jersey cows, which had not such power of converting carotene into vitamin A, the colour of the milk remained lighter.

Dr. D. KENT-JONES asked what sort of agreement there was among analysts when dealing with alfalfa grasses, as he had found that the variation between results obtained by different analysts was very large. As regards the feeding of carotene, when it was oxidised, as it was in bleached flour, did one destroy the carotene from the point of view of vitamin A? It was generally agreed that this was so, but papers had been published in which the contrary was stated.

Mr. A. L. BACHARACH referred to the shortage of Merck's alumina; the suggested procedure of recovering it after use might involve destruction of its adsorbent properties. The excellence of Merck's product might lie in the presence of a certain amount of the so-called "fibrous" alumina (*Wislicenus*). There had recently been marked improvement in the adsorbent alumina made in this country, and probably at least two were quite satisfactory. In spite of the importance of the conversion of carotene into vitamin A, little was known about it. "Carotinase" might exist in living tissues, but it had only so far been seen in text-books. The conversion ratios of carotene and vitamin A were singularly constant, according to Guilbert, for different species, but varied very much with intake of carotene,

which might have in mammals one-third the molar activity of vitamin A at low doses and only 1/10th at high doses. If Dr. Morton was right in suggesting that there was no reason why carotene should break down at any one double bond more than another, a very complex series of changes must be involved in the conversion, and it was not surprising that no single conversion factor could be applied.

Dr. R. L. EDWARDS, referring to the yellow colour of the carotene solution, said that in the work of the Committee on carotene determination in dried grass mentioned by Mr. Seaber, readings were taken by about twenty-five observers on solutions of dichromate (0.025 per cent.) and on carotene (0.10, 0.20, 0.25 mg. per 100 ml.). Each observer calculated his own carotene concentration equivalent to 0.025 per cent. dichromate from these readings. If there were no inherent disadvantage in using Lovibond glasses, one would expect a standard deviation from the mean of this carotene equivalent involving two readings with their combined errors, to be greater than that of either of the separate readings. Actually, it was less than either. Expressed as percentage of the mean, Lovibond reading on 0.025 per cent. dichromate, s.d. 11.5 per cent.; Lovibond reading on carotene 0.10 mg. per 100 ml., s.d. 14 per cent.; 0.20–14 per cent.; 0.25–21 per cent. Carotene equivalent to 0.025 per cent. dichromate, s.d. 5.5 per cent. It seemed that one obtained better agreement between laboratories if one used the Lovibond Tintometer simply as a comparator for dichromate and carotene. In connection with the theory of unsymmetrical splitting of carotene, it was interesting to carry the speculation a little further to include α - and γ -carotenes, which were supposed to be only half as active as β -carotene. This could be explained if, whatever the mechanism involved, splitting of the carotene molecule occurred with equal frequency on each side of the centre, regardless of the nature of the end groups. Then 50 per cent. of the α -carotene molecules would give β -ionone rings with chains long enough to form vitamin A, whilst the other 50 per cent., having β -ionone with short chains, would be inactive.

Mr. D. J. CAMPBELL referred to a sample of magnesia, apparently unactivated, which he had found to be quite suitable for the separation of carotenoids, and which could be used with 3 per cent. acetone. It seemed to have been established that carotene was most easily available to the animal when fed in oil, and he would like to know if any work had been done on feeding carotene in oil-cake. The availability of carotene was very important. With regard to the Lovibond Tintometer, he thought that errors were often really due to the faulty colour sense of observers. For carotene estimations a personal calibration was necessary. As regards the concentration of methyl alcohol for xanthophyll removal, he had recently made experiments with authentic carotene, and the exact concentration between 85 and 98.5 per cent. alcohol did not seem to make any difference; the stronger the alcohol the more petrol passed into it, but provided that enough free petrol was present, no carotene whatever was removed, even when the alcohol was 98.5 per cent. in strength.

Dr. MORTON, replying, said that Lovibond glasses of recent manufacture left little to be desired, although those of an old set, dated 1881, which he had examined, were far from satisfactory. In reply to Mr. Bacharach, he pointed out that he had reviewed the present situation more in the hope of making clear the difficulties than of sponsoring any particular theory. There was much to be said for buying and selling on the basis of the chemical content in terms of carotene and vitamin A. For many products, although not perhaps all, the time had gone for biological tests, since even a crude physical assay for vitamin A and carotene was usually more significant than any but the most ambitious biological tests. The great impetus to vitamin A studies had come from biochemical research, and he wished to see the skill of the biochemist released from routine standardisation and applied to the fundamental problems which could be solved by no other type

of worker. Concerning chromatographic analysis he remarked that his own experience bore out the view that experiments on a very small scale gave the best analytical results.

Mr. SEABER, replying, said that his own experience of the use of the Lovibond Tintometer had been rather unfortunate. He had some difficulty sometimes with the yellow colours. He had found that many people said that they got very good results provided that the Lovibond number was not too large. He was very interested to hear that Dr. Kon confirmed his own findings relating to the non-carotene substances usually included as carotene. With regard to substitutes for Merck's alumina, he had tried magnesia; it had recently been prepared in such a way that it adsorbed the xanthophyll and not the carotene. With regard to methyl alcohol, they had found that when they used it in 85 per cent. strength in ordinary work they obtained lower results than with 90 per cent. Dr. Kent-Jones had asked what sort of variations chemists obtained in estimating carotene. Sometimes they were enormous; for example, from 150 to 350 parts per million. That was why the Grass Driers' Association had formed the Committee to which he had referred. On the other hand, in his own laboratory, they found they could get very close agreement in their results.

The Separation of Molybdenum from Tin and from Sulphur

By D. A. LAMBIE, B.Sc., A.I.C., AND W. R. SCHOELLER, Ph.D., F.I.C.

In the course of a revision of the analytical chemistry of molybdenum, we found it advisable to re-investigate the separation of that element from tin and from sulphur (in the form of sulphate ion). The results of our work are recorded below.

I. SEPARATION OF MOLYBDENUM FROM TIN.—This separation does not appear to have been thoroughly studied, although the following methods have been proposed: (1) precipitation of molybdenum by hydrogen sulphide from oxalic acid solution; (2) precipitation of molybdenum by hydrogen sulphide from hydrofluoric acid solution; (3) volatilisation of stannic chloride by the action of dry hydrogen chloride upon a strong sulphuric acid solution at 200° C.; (4) for small amounts of tin, precipitation of hydrated stannic oxide on ferric hydroxide—a method employed for collecting minute quantities of a number of elements (phosphorus, arsenic, antimony, bismuth, vanadium, selenium, tellurium).

At the outset we decided to investigate methods (1) and (4), so as to avoid, if possible, the use of platinum vessels (method 2) or special all-glass apparatus (method 3).

Precipitation of Molybdenum Sulphide from Oxalic Acid Solution is an adaptation of Clarke's method for the separation of trivalent antimony from quadrivalent tin.¹ Since the quantitative course of the separation of molybdenum from tin by this reaction does not appear to have been investigated, we conducted a series of experiments with the following procedure, based upon a modification of Clarke's method.²

Known amounts of tin and molybdenum trioxide were dissolved in *aqua regia*, 15 ml. of 20 per cent. tartaric acid solution were added, and the solution was

neutralised with 20 per cent. sodium hydroxide solution; another 20 ml. of the alkali were then added, and the liquid was saturated with hydrogen sulphide. The solution of the thio-salts was boiled and cautiously treated with 15 g. of oxalic acid dissolved in a little hot water, after which hydrogen sulphide was again passed for 15 minutes. The precipitate was collected, washed with hot 1 per cent. oxalic acid solution containing hydrogen sulphide, and dissolved in nitric acid and bromine; the solution was then neutralised with ammonia prior to precipitation of lead molybdate. In all the experiments but one, the neutralisation with ammonia produced a precipitate of stannic acid, proving the separation to have been incomplete. In one test (on 0.1544 g. Sn and 0.0701 g. Mo), the stannic acid thus produced was collected and ignited, yielding 0.0072 g. of impure oxide contaminated with molybdenum. The test separation in which no stannic acid was precipitated on neutralisation gave the following result:

Taken, 0.0300 g. of Sn, 0.1000 g. of Mo; found 0.1008 g. of Mo.

In a second series of tests the hot solution of the thio-salts, obtained as above, was poured into the hot oxalic acid solution in an endeavour to obtain a molybdenum sulphide precipitate free from tin; but even so, a satisfactory separation was not achieved except with small quantities of tin.

It is possible, indeed probable, that a quantitative separation could be brought about by re-treatment of the molybdenum sulphide, the precipitate being dissolved in sodium sulphide and the solution poured into hot oxalic acid solution. The combined filtrates containing the tin are, however, liable to be contaminated with a little molybdenum, the recovery of which would prove troublesome and constitute a serious drawback to the method.

These considerations induced us to suspend further work on the sulphide method, and to investigate the possibilities of the process about to be described.

Precipitation of Hydrated Stannic Oxide on Ferric Hydroxide.—As far as we know, the application of this procedure to the quantitative separation of molybdenum from tin in any proportions has not been investigated. Ferric iron can be separated from molybdenum by double precipitation with ammonia, the hot feebly acid solution of the metals being poured during agitation into almost boiling dilute (1 : 1) ammonia. If stannic salt also is present, the tin is precipitated together with the iron, the molybdenum passing into the filtrate as ammonium molybdate. We succeeded in working out conditions under which a quantitative separation of molybdenum from substantial, as well as from small, quantities of tin is achieved, but not without numerous experiments bearing upon the ammonia concentration, the admissible maximum quantities of tin and molybdenum, and the most suitable ratio of iron to tin.

In the final series of tests the results given below were obtained by the following procedure, which we recommend for the separation of the two elements:—The weighed quantities of tin and molybdic oxide were dissolved in 10 ml. of strong hydrochloric acid, and 3 ml. of strong nitric acid; the quantities taken were adjusted to a maximum amount of 0.3 g. of (tin *plus* molybdenum). The resulting solution was treated with enough ferric chloride solution to provide iron equal in weight to the tin present, and diluted to 100 ml. The liquid was heated nearly to boiling, and poured during vigorous agitation into 100 ml. of dilute ammonia

(20 ml. of ammonia of sp.gr. 0.88, and 80 ml. of water already at the boiling-point), boiled for half-a-minute, and allowed to settle. The precipitate was collected on a close-textured filter, washed with 3 per cent. ammonium nitrate solution, returned to the beaker with a jet of water, and dissolved by heating with 10 ml. of strong hydrochloric acid (total volume 50 to 60 ml.). The resulting solution was then diluted to 100 ml., and the operation described above was repeated, the same filter being employed.

The combined filtrates were concentrated to about 200 ml., and the molybdenum was determined in the usual manner as lead molybdate. The tin was determined in the precipitate by the gravimetric method outlined below. The results were well within the limit of experimental error.

| Exp. | Taken | | Found | | Error | |
|------|----------|----------|----------|----------|----------|----------|
| | Sn g. | Mo g. | Sn g. | Mo g. | Sn g. | Mo g. |
| 1 | 0.2930 | 0.0068 | not det. | 0.0065 | — | -0.0003 |
| 2 | 0.2092 | 0.0685 | 0.2095 | 0.0685 | +0.0003 | 0.0000 |
| 3 | 0.1029 | 0.1347 | 0.1033 | 0.1352 | +0.0004 | +0.0005 |
| 4 | 0.1515 | 0.1486 | 0.1511 | 0.1484 | -0.0004 | -0.0002 |
| 5 | 0.0243 | 0.2676 | 0.0243 | 0.2673 | 0.0000 | -0.0003 |
| 6 | 0.0134 | 0.2813 | 0.0134 | 0.2822 | 0.0000 | +0.0009 |

Gravimetric Determination of Tin.—While the separation of molybdenum from tin is under discussion, a few remarks may be usefully devoted to the determination of tin. The iodimetric titration is subject to interference by atmospheric and dissolved oxygen,³ and the precautions to overcome this detract from the practical value of the process for occasional determinations. For these, precipitation of the tin as sulphide and ignition of the precipitate to oxide is really less troublesome, provided that other members of the hydrogen-sulphide group are absent, or have been eliminated by the usual methods, *e.g.* by means of sodium sulphide or *ferrum redactum*. In the procedure described below, which has been used for some time in actual practice with very satisfactory results, the stannic sulphide precipitate is dense and usually granular, filters well, especially if mixed with a little filter-fibre, and readily attains constant weight on ignition.

Insoluble material is fused with sodium peroxide or hydroxide as usual. The melt is disintegrated with hot water, and transferred to a 150- to 400-ml. beaker, and the crucible is warmed with dilute sulphuric acid till clean. The acid is transferred to the beaker, the contents of which are treated with 1 : 1 sulphuric acid until an excess of 10 to 30 ml. is present. The liquid is then evaporated until strong white fumes are evolved.

Precipitates (such as the mixed precipitate of hydrated stannic and ferric oxides obtained in the separation procedure under discussion) are placed in a beaker together with the filter-paper, and heated with strong sulphuric acid; nitric acid is cautiously added until all the organic matter is destroyed, after which the liquid is heated more strongly until dense sulphuric acid fumes are given off.

The cold acid is diluted with an equal volume of water, followed by 20 to 50 ml. of 20 per cent. ammonium chloride solution. The liquid is digested at

gentle heat until any anhydrous nickel or ferric sulphate has dissolved, then diluted, treated with creamed filter-fibre to entangle gelatinous silica, and filtered through a pad of filter-pulp into a 250- to 600-ml. beaker. The filter-pad is washed first with hot 0.5 *N* hydrochloric acid, and then with water.

The filtrate is strongly diluted (200 to 400 ml.) and saturated with hydrogen sulphide; addition of a little creamed filter-fibre causes rapid flocculation of small quantities of stannic sulphide. The precipitate is allowed to settle, collected, washed thoroughly with one per cent. sulphuric acid containing hydrogen sulphide, ignited wet in a tared porcelain crucible, first on an asbestos mat until the paper has charred, then a little more strongly until the carbon has been oxidised, finally at a bright red heat, and weighed as SnO_2 (cf. A. T. Etheridge, *ANALYST*, 1924, 49, 371).

II. SEPARATION OF MOLYBDENUM FROM SULPHUR.—In practice this resolves itself into the separation of molybdate from sulphate, or rather (since the two elements are not usually determined in one and the same solution) into: (a) determination of molybdate in presence of sulphate; (b) determination of sulphate in presence of molybdate.

(a) The determination of molybdate as lead molybdate in presence of sulphate by a single precipitation (dropwise addition of a small excess of lead salt to a solution containing a large excess of ammonium acetate and chloride)⁴ offers no special difficulties, re-treatment of the precipitate by solution in hydrochloric acid and re-precipitation by means of a large excess of ammonium acetate being necessary only with greatly preponderating quantities of sulphate.

(b) The determination of sulphate as barium sulphate in presence of molybdate, which is of practical interest in the analysis of molybdenum ores, is a subject concerning which very little information is available. We re-investigated a procedure described by Schoeller and Powell,⁵ consisting in dropwise addition of 10 per cent. barium chloride reagent to the boiling solution containing 5 per cent. by volume of strong hydrochloric acid. The pronounced solubility of barium sulphate at this acid concentration was found to be more than counteracted by occlusion of molybdenum in the precipitate, with the result that positive errors were incurred. Only one of our test separations gave a seemingly correct result, yet the barium sulphate was obviously contaminated with molybdenum, being yellowish-green in colour; hence the agreement between the calculated and observed values was due to a compensation of errors. The method was rejected as untrustworthy.

Reduction of the molybdic acid to a lower compound prior to precipitation of the solution with barium chloride was tried next, but proved to lead to heavier co-precipitation of molybdenum than the preceding method. We reached the conclusion that, owing to the marked adsorptive power of barium sulphate, elimination of the molybdate prior to precipitation of the sulphate with barium chloride was the correct solution of the problem. In the process described below, the molybdate is precipitated with a slight deficiency of lead acetate in presence of ammonium acetate, and the ammonium salts (which would increase the solubility of barium sulphate) are removed from the filtrate by evaporation with *aqua regia*. The minute amount of molybdate left in the solution is much too small to interfere.

Two reagents are required: (1) a lead solution containing 40 g. of lead acetate and 10 ml. of glacial acetic acid per litre, and (2) a molybdate solution containing 1.86 g. of ammonium molybdate per 100 ml.; the two solutions are approximately equivalent.

The solution, containing not more than 0.3 g. of molybdenum and 0.2 g. of sulphur (the proportions in which the elements occur in molybdenite) as molybdate and sulphate, respectively, is neutralised with ammonia to methyl orange, treated with 5 ml. of glacial acetic acid and 12.5 g. of ammonium acetate, diluted to 200 ml., heated to boiling, and titrated dropwise with the lead solution until a drop fails to give a colour reaction with a freshly-prepared 0.5 per cent. tannin solution on a porcelain tile; the molybdate solution is then added from a 1-ml. pipette until the tannin reaction just re-appears. The suspension is boiled for 15 to 20 minutes and the precipitate is allowed to settle, filtered off, and washed with 2 per cent. ammonium acetate solution. For accurate work the precipitate should be returned to the beaker and dissolved by heating with dilute hydrochloric acid. The boiling solution is treated with 1:1 ammonia until a slight permanent turbidity is produced, then with 10 g. of ammonium acetate, and boiled for 15 to 20 minutes. The precipitate is collected, washed as before, and discarded. The combined filtrates are boiled down and finally evaporated on a water-bath with hydrochloric acid for the expulsion of the acetic acid, after which the beaker is covered and excess of strong nitric acid is added, the heating being continued until all effervescence has ceased. The cover and sides of the beaker are then rinsed down, and the nitric acid is removed in the usual manner by evaporation to dryness and two successive evaporations with hydrochloric acid. The sulphuric acid is then precipitated in the usual manner with barium chloride. A blank test on the reagents used should not be omitted.

The results of four test analyses by this process are given below. Pure molybdic oxide dissolved in dilute ammonia was added to measured amounts of a standard solution of ammonium sulphate (Analar), the sulphate-content of which had been determined gravimetrically. In Exp. 1, the lead molybdate was not re-precipitated, the single treatment resulting in a fairly appreciable negative error. In Exp. 2, the lead molybdate was re-treated, and the two filtrates were worked up separately for sulphur. Exps. 3 and 4 were conducted as described above, the lead molybdate being re-treated and the filtrates combined. The blank was equivalent to 0.0007 g. of sulphur.

| Exp. | Taken | | S found | | | Error S g. |
|------|----------|---------|-----------------|-----------------|-------------|------------------|
| | Mo g. | S g. | 1st pptn. g. | 2nd pptn. g. | Total g. | |
| 1 | 0.300 | 0.1946 | 0.1930 | — | 0.1930 | -0.0016 |
| 2 | 0.300 | 0.1934 | 0.1926 | 0.0010 | 0.1936 | +0.0002 |
| 3 | 0.300 | 0.1934 | — | — | 0.1927 | -0.0007 |
| 4 | 0.300 | 0.1934 | — | — | 0.1928 | -0.0006 |

SUMMARY.—Certain methods for the separation of molybdenum from tin and from sulphur are criticised, and improved processes for the two separation cases are presented. A convenient procedure for the gravimetric determination of tin is described.

REFERENCES

1. F. W. Clarke, *Chem. News*, 1870, **21**, 124.
2. F. P. Treadwell and W. T. Hall, "*Analytical Chemistry*," Vol. II, 8th Ed., 1935, 238.
3. F. L. Okell and J. Lumsden, *ANALYST*, 1935, **60**, 803.
4. W. R. Schoeller and A. R. Powell, "*The Analysis of Minerals and Ores of the Rarer Elements*," London, 1919, p. 164 (New Edition in the press); J. P. Bonardi in *Bull.* 212, U.S. Bureau of Mines, 1923, p. 102; Arrington and Rice, *Id.*, R.I. 3441, 1939, pp. 39-59.
5. W. R. Schoeller and A. R. Powell, *op. cit.*, p. 160.

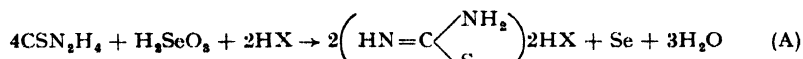
February, 1940

The Determination of Thiourea by Oxidation with Selenious Acid

BY A. E. A. WERNER, PH.D., M.Sc., A.I.C.

PREVIOUS methods^{1,2} outlined for the determination of thiourea depend mostly on oxidation of the thiourea molecule by iodine, and require to be carried out under carefully prescribed conditions in order to yield consistently accurate results.

The present method is based on the straightforward reaction which takes place in acid solution between thiourea and selenious acid in accordance with the following equation:



i.e. one molecule of selenious acid oxidises four molecules of thiourea with the production of a salt of bis-(amino-imino-methyl)-disulphide. Preliminary experiments have shown that the reaction proceeds quantitatively over a wide range of thiourea concentrations, and it therefore offers the basis for a rapid and accurate method for the determination of thiourea in solution.

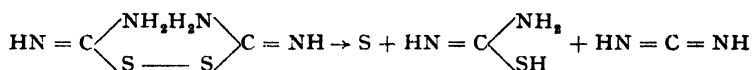
A known excess of a standardised solution of selenious acid is added to the thiourea solution, and the amount of selenious acid left unused is determined either iodimetrically or by direct titration with standard alkali. Since selenious acid reacts with potassium iodide in acid solution with the liberation of iodine according to the following equation:



this reaction affords an admirable method for its determination, the liberated iodine being determined in the usual manner with *N*/10 sodium thiosulphate solution; the end-point is very sharp, and the violet colour of colloidal selenium does not interfere, as the selenium precipitate is readily coagulated, on shaking, to a bulky precipitate, leaving a colourless supernatant liquid (*cf.* Muthman and Schäfer³). Selenious acid can also be determined by direct titration with standard alkali, sodium alizarinate being used as indicator; a sharp colour-change from yellow to red occurs at a point corresponding with the formation of sodium hydrogen selenite. However, the iodimetric method is preferable owing to its greater accuracy, as the titration figure is four times as great for the same normality.

REAGENTS REQUIRED.—Solution of $M/5$ selenious acid in $N/5$ sulphuric acid; $N/10$ sodium thiosulphate solution; 5 per cent. potassium iodide solution; $N/20$ sodium hydroxide solution. The solution of selenious acid is made by dissolving 3.22 g. of selenious acid in 200 ml. of water, to which 50 ml. of N sulphuric acid have been added; this solution is then accurately standardised by adding 2 ml. to excess of potassium iodide solution and determining the liberated iodine by titration with $N/10$ sodium thiosulphate solution, or alternatively by direct titration with $N/10$ sodium hydroxide solution.

METHOD.—The following technique has been found to yield consistent and accurate results. Ten ml. or 5 ml. (depending on the concentration) of the unknown thiourea solution are added to 10 ml. of the standard selenious acid solution. After the fine precipitate of selenium has been coagulated by addition of sodium chloride, the solution is filtered through a sintered glass filter, and the clear filtrate is made up to 100 ml. with water. Twenty-five ml. portions of the filtrate are added to excess of potassium iodide solution, a few drops of sulphuric acid are added, and the iodine liberated is titrated with $N/10$ sodium thiosulphate solution. The filtrate should not be left standing before the titration, because in the now dilute acid solution the salt of bis-(amino-imino-methyl)-disulphide slowly decomposes with precipitation of sulphur and generation of more thiourea, which would react further with the selenious acid. This decomposition is represented by the following equation:



To illustrate the degree of accuracy obtainable by this method some typical results are included in the following tables.

TABLE I

RESULTS OF THIOUREA DETERMINATION. IODIMETRIC METHOD

| Volume of thiourea soln. added ml. | Volume of selenious acid ml. | Thiosulphate titre per 25 ml. ml. | Thiourea | |
|---|---------------------------------------|--|--------------------|--------------------------|
| | | | Found Per Cent. | Theoretical Per Cent. |
| 0 | 10 | 10.60 | — | — |
| 10 | 10 | 8.95 | 0.50 | 0.50 |
| | | 9.00 | 0.48 | |
| 10 | 10 | 8.00 | 0.79 | 0.80 |
| | | 7.95 | 0.80 | |
| 10 | 10 | 7.40 | 0.97 | 1.00 |
| | | 7.35 | 0.99 | |
| 5 | 10 | 8.10 | 1.52 | 1.50 |
| | | 8.15 | 1.49 | |
| 5 | 10 | 8.00 | 1.95 | 2.00 |
| | | 7.90 | 2.00 | |
| 5 | 10 | 3.70 | 4.19 | 4.00 |
| | | 3.75 | 4.15 | |

From equations (A) and (B) it follows that 1000 ml. of $N/10$ sodium thiosulphate correspond with 7.6 g. of thiourea. Therefore, according as 10 ml. or 5 ml. of the thiourea solution are used, and when 25 ml. of the filtrate are taken for the titration, the percentage of thiourea is given by the expression:

$$0.0076 \times (x - y) \times 40 \quad \text{or} \quad 0.0076 \times (x - y) \times 80$$

where x = titre without the addition of thiourea; y = titre after the addition of thiourea.

TABLE II

RESULTS OF THIOUREA DETERMINATIONS. DIRECT ACID TITRATION

| Volume of thiourea soln. added ml. | Volume of selenious acid ml. | N/20 NaOH, titre per 25 ml. ml. | Thiourea | |
|---|---------------------------------------|--|--------------------|--------------------------|
| | | | Found Per Cent. | Theoretical Per Cent. |
| 0 | 10 | 14.90 | — | — |
| 10 | 10 | 13.60 | 0.79 | 0.80 |
| | | 13.70 | 0.76 | |
| 10 | 10 | 13.00 | 1.15 | 1.10 |
| | | 12.90 | 1.20 | |
| 5 | 10 | 13.70 | 1.46 | 1.50 |
| | | 13.65 | 1.52 | |
| 5 | 10 | 13.30 | 1.95 | 2.00 |
| | | 13.35 | 1.90 | |

From equation (A) it follows that 1000 ml. of $N/10$ sodium hydroxide are equivalent to 30.4 g. of thiourea. Therefore, according as 10 ml. or 5 ml. of the thiourea solution are taken, and when 25 ml. of the filtrate are used for each titration, the percentage of thiourea is given by the expression:

$$0.0304 \times (X - Y) \times 20 \quad \text{or} \quad 0.0304 \times (X - Y) \times 40$$

where X = $N/20$ NaOH titre without addition of thiourea; Y = $N/20$ NaOH titre after addition of thiourea.

I wish to express my thanks to Dr. E. A. Werner for his many valuable suggestions.

REFERENCES

1. E. A. Werner, *J. Chem. Soc.*, 1903, 83, 1.
2. C. Mahr, *Z. anal. Chem.*, 1939, 117, 91.
3. C. Muthman and W. Schäfer, *Ber.*, 1893, 26, 1008.

January, 1940

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

A RAPID STABILITY TEST FOR SEWAGE EFFLUENTS

THE methylene-blue test for relative stability would be of greater value if it took less time to complete. Many workers have attempted to modify the procedure with the object of reducing the time, but most of the attempts have succeeded only at the expense of the simplicity of the original test. Since methylene blue is only one of a large number of oxidation-reduction indicators, there appears to be no particular reason for retaining it in preference to other indicators, especially when its varied toxic effects—as shown, for example, by the work of Yudkin¹—are taken into account. Remy² has compared the sensitivity of dichlorophenol-indophenol with that of methylene blue and has found it to react more easily and with greater certainty in presence of putrescible sulphur compounds.

While investigating the adsorption and reduction of dyestuffs by activated sludge, Dickinson³ found that methylene green (nitro-methylene blue) is reduced very rapidly and that the reaction is not inhibited by the presence of air. This reaction has been studied in some detail, and the rate of reduction at constant temperature has been found to correspond with the equation $t = aC^n$, where t is the time taken to reduce a quantity C , and a and n are constants. The quantity n appears to be constant for the reaction, while a varies with the sludge. Since all sewage effluents contain particles of sludge in suspension, or the active principles thereof, it was thought profitable to explore the possibilities of methylene green as a substitute for methylene blue. The following method was eventually adopted:—Methylene green solution (0.2 ml. of an aqueous solution containing 2 g. per litre), 0.2 ml. of an aqueous solution of methyl orange (1 g. per litre), and 1 ml. of $M/15$ potassium dihydrogen phosphate solution are measured into a 4-oz. bottle of the type usually used for the methylene-blue test. The bottle is then filled with the sample under examination and incubated at 80° F. The time taken for the solution to change colour from green to yellow is measured. If a "blank" is used for comparison, the end-point is more clearly seen. The methylene green was prepared by the method given by Cumming, Hopper and Wheeler.⁴

The methyl orange was included to render the end-point more distinct; it has no other influence on the test. The phosphate buffer is essential, as in its absence the pH of the sample rises, the methylene green changes colour, and the reduction is inhibited. The following table summarises the results obtained:

| Methylene green reduced in: | B.O.D.* | No. of successful tests | No. of exceptions |
|---------------------------------|---------------|-------------------------|-------------------|
| Less than $\frac{1}{2}$ hour .. | >4.0 | 33† | 2 |
| $\frac{1}{2}$ to 1 hour | 3.0 to 5.0 | 13 | 0 |
| 1 to 8 hours | 2.0 to 4.0 | 12 | 3 |
| More than 8 hours .. | Less than 2.0 | 18 | 1 |
| | Total: | 76 | 6 |

* B.O.D.: Biochemical oxygen demand, parts per 100,000.

† Only 4 had a B.O.D. less than 5.0.

The B.O.D.'s were measured variously by the Winkler azide method in 5 days at 65° F., and by the ordinary Winkler method in 3 days at 80° F. Of a number of effluents from a percolating filter, all took at least 30 hours to decolorise the methylene green, and all had a B.O.D. of less than 2.0. Effluents from contact beds reacted with much less certainty than any of the other samples examined.

It is important to point out that the reduction of methylene green is essentially different from the reduction of methylene blue, for whereas the latter is employed as an indicator, the former is used as a reagent, and its reduction does not indicate the exhaustion of dissolved oxygen in the sample.

We have found that the methylene-green test is a useful adjunct to routine analysis, especially when it is desired to form a provisional opinion quickly. It has also proved useful in determining the amount of dilution water to be added to doubtful samples when preparing the B.O.D. tests.

D. DICKINSON

E. MCGREGOR WEIR

REFERENCES

1. G. Yudkin, *Biochem. J.*, 1933, **27**, 1849.
2. E. Remy, *Arch. Hyg. Berl.*, 1937, **119**, 141.
3. D. Dickinson, *J. Soc. Chem. Ind.*, 1939, **58**, 199.
4. W. C. Cumming, J. V. Hopper and T. S. Wheeler, "Systematic Organic Chemistry," Constable, London, 1923, p. 382.

March 30th, 1940

Department of Scientific and Industrial Research

METHODS FOR THE DETECTION OF TOXIC GASES IN INDUSTRY

PHOSGENE*

OCCURRENCE.—Phosgene (carbonyl chloride) may be encountered in dangerous concentrations in works manufacturing organic chemical and pharmaceutical products, and, in particular, dyestuffs; also as a decomposition product of carbon tetrachloride (used, for example, in fire extinguishers).

POISONOUS EFFECTS.—Phosgene causes severe damage to the alveoli of the lungs, and this is followed by pulmonary oedema, resulting in asphyxiation. The immediate symptoms produced by even a fatal dose may be relatively mild—a little coughing, tightness of the chest and lachrymation—but, after apparently recovering, the patient may become progressively and acutely ill, owing to the injury to the lungs. An atmosphere containing 1 part (by vol.) of the gas in 6000 may cause lung injuries in 2 minutes, 1 part in 30,000 is very dangerous, and 1 part in 200,000 is probably fatal in 30 minutes. The maximum permissible concentration for a prolonged period is about 1 in 1,000,000 (0.004 mg. per litre).

DETECTION.—The yellow or orange stain produced by phosgene on test-paper containing diphenylamine and *p*-dimethylaminobenzaldehyde has been adopted as the standard test for the detection of the gas in industry. The test, which is capable of detecting about 1 part of phosgene in 1,000,000 of air, has been made quantitative by drawing known volumes of the atmosphere through a definite area of the test-paper by means of a hand-pump of specified capacity, and noting the number of strokes required to produce stains of certain intensity; the corresponding concentration is then found by reference to standard stains on a colour chart supplied with the pamphlet. The stains produced by phosgene are only transient, and it should also be noted that the test-papers are sensitive also to chlorine or hydrogen chloride. To remove traces of these gases the atmosphere is passed through a guard-tube containing pumice granules impregnated with sodium thio-sulphate before it comes in contact with the test-paper.

* Leaflet No. 8. H.M. Stationery Office, York House, Kingsway, London, W.C.2. 1939. Price 2s. 6d. net. Further copies of the Standard Stains, price 2s.; by post 2s. 3d.

Western Australia

ANNUAL REPORT OF THE CHEMICAL BRANCH, MINES DEPARTMENT, FOR 1938

THE Chemical Branch of the Mines Department, which is under the direction of the Government Analyst (Dr. E. S. Simpson), undertakes the chemical work of all the Government Departments. The total number of samples examined during the year was 6036, and these are classified into three sections: (1) food, drugs and toxicology (893), (2) mineralogy and geo-chemistry (2987) and (3) agriculture and water supply (2156). Of the 120 samples of food, 32 were below standard.

The Food and Drugs Advisory Committee dealt with the following questions among others:

Preservatives in Butter.—The regulation dealing with butter was amended by withdrawing the permission to use any preservative in it other than salt. Previously boric acid to the extent of 0.3 per cent. was allowed.

Labelling of Baking Powder.—A request that the words "acid phosphate" should be removed from labels on baking powder containing this substance was refused. This was on account of the large amount of fluoride that had been found in certain commercial acid phosphates.

n-Butyl-p-hydroxybenzoate as Preservative.—Pending further information, it was decided not to add this substance (which is sold under several trade names) to the list of permitted preservatives.

Arsenic and Lead Limits for Fresh Vegetables.—"Fresh vegetables" were added to the list of foodstuffs in which the amount of arsenic must not exceed the equivalent of 1-100th grain of As_2O_3 per lb., and that of lead must not exceed 1-7th grain per lb.

Points of interest to which the Report directs attention include the following: "SWEET MANGO CHUTNEY."—A sample bearing this label was found to contain only traces, if any, of mango, the main constituents being apple, onion and tomato, with raisins, currants, cloves, salt, etc.

VITAMIN C CONTENTS OF WEST AUSTRALIAN FRUITS.—The very high proportions found in a large yellow guava (1.10 mg.) and a papaw (0.98 mg. per g.) are noteworthy. Figures for other fruits agreed closely with those found in other parts of the world, the richest common fruits being orange, mandarin, lemon and "banana passion fruit" (also known as taxonia).

INDUSTRIAL POISONING WITH LEAD AND ARSENIC.—An investigation was made of the liability of men engaged in the Wiluma gold mines to poisoning with lead and arsenic. Eighty-nine specimens of urine and 29 specimens of hair and nails were examined, and the following range of figures was recorded:

| | | Lead as Pb p.p.m. | Arsenic as As_2O_3 p.p.m. |
|-------|-------|----------------------|--------------------------------|
| Urine | | nil to 0.75 | 0.07 to 3.75 |
| Hair | | — | nil to 3880 |
| Nails | | — | 9 to 1250 |

In most cases in which high results were obtained the patients showed symptoms of poisoning with lead or arsenic or both.

DETERMINATION OF LEAD IN URINE.—The following rapid method has been found to give accurate results. Organic matter is destroyed with the minimum amount of sulphuric and nitric acid plus a little perchloric acid, and the dithizone method of extraction is employed in presence of ammonium citrate and potassium cyanide at pH 9.0 to 9.5. The excess of dithizone is removed, and the red colour of the lead complex is compared with the colours obtained under similar conditions with standard lead solutions.

LEAD COMPOUNDS IN CELLULOSE SPRAY PAINTS.—The presence of considerable quantities of lead compounds was proved in two kinds of lacquers used in the spray painting of motor vehicles. Tests of the respirators worn by the workmen showed that they were effective in retaining lead, but constant watching is necessary to ensure that the spray-painting regulations are observed, and that the respirators fit closely and are frequently cleaned and renewed.

POISONOUS FUMES FROM AN ARC LIGHT.—As the result of a complaint by an association of employees an investigation was made of the conditions in the operating room of a cinema where blue flame carbons are used. During the working of the arc, and especially on making contact, white fumes arise from the incandescent zone and form a white deposit in the chamber unless conducted away by an efficient draught. Analysis showed the carbons to be copper-sheathed and to contain a core of cerium fluoride cemented with a silicate composition. The fumes were found to consist of cerium compounds with some fluorine and copper. The symptoms experienced by the operatives were consistent with the ingestion of small amounts of copper; in view also of the known toxicity of fluorine compounds, recommendations were made for improved ventilation and frequent removal of any deposit.

PINE SEEDLINGS DISEASE AND SILVER.—A number of pine seedlings and needles were examined spectrographically to see if any mineral deficiency could be detected. Interesting results were obtained with seedlings from certain plantations where a disease characterised by stunted growth, discoloration and altered pose of the needles was manifest. Spectrographic analysis showed larger amounts of silver in the unhealthy than in the healthy pines.

POISONOUS PRINCIPLE OF ZAMIA FRUIT.—Parts of the fruit of the zamia plant (*Macrozamia Fraseri*, Miq.), common locally, are poisonous to cattle, producing gastro-intestinal irritation. The fruit consists of a hard seed or nut containing a starchy endosperm surrounded, when ripe and fresh, by a thin fleshy layer of mesocarp with an orange-red epicarp. Only the seeds were found to be poisonous—a toxic principle not precipitated by lead acetate, and not extracted by immiscible solvents, being present. The fruit pulp surrounding the seeds contained 14 per cent. of a bright orange-coloured oil, which appears to contain a considerable amount of carotene and closely resembles palm oil in its physical and chemical constants. Further work is in progress.

STANDARD FOR IRRIGATION PURPOSES.—Twenty-four samples of waters have been examined with the object of fixing a standard for irrigation purposes. It was found that waters containing up to 258 grains of total salts per gallon were being used for the irrigation of grape vines, and waters containing up to 276 grains for lucerne, rhubarb, cabbage, and cauliflower cultivation. The sodium chloride content of these waters was usually 75 per cent. or more of the total salts. These figures are much higher than the usually accepted standards for irrigation waters.

COPPER IN APPLE LEAVES.—Samples of leaves from apple trees were examined. In some the soil round the trees had been treated with copper sulphate, and in others copper sulphate had been injected into the trees. The copper found in the leaves ranged from less than 1 p.p.m. to 5.1 p.p.m. in the dried leaves.

Coal Mines Act, 1911

PRECAUTIONS AGAINST COAL DUST*

THIS pamphlet is in three Parts: I The Coal Mines General Regulations (Precautions against Coal Dust), 1939, showing certain modifications made temporarily in view of difficulties created by the war (S.R. & O., 1939, No. 1804). II. The Mine Dust Analysis Order, 1939, prescribing the methods to be followed in analysing samples of mine dust collected in accordance with the Regulations. III. A Memorandum for the information of chemists who are called upon to analyse samples of mine dust for the purpose of the Regulations.

PART I. PRECAUTIONS AGAINST COAL DUST.—The Regulations provide that in every part of the "road" of the mine which is accessible the dust that can be raised into the air shall contain, when tested in the manner prescribed, not less than the percentage of incombustible matter set out in the Schedule to the Regulations (*infra*) according to the volatile matter content of the coal.

The percentage of incombustible matter means the actual percentage of incombustible matter (including moisture) contained in the dust, plus any percentage allowance permitted on account of any of the incombustible matter which is of superior efficacy as compared with ordinary shale dust. The permitted percentage allowance, if any, shall be calculated from the analysis of the dust in the appropriate manner prescribed by the Board of Trade. (*Note.*—For the present no allowances under this provision are to be permitted.)

The volatile matter content of the coal means the average volatile matter content calculated on an ash-free dry basis of the seam of coal worked through the road (or, if more than one seam is worked, of that seam which has the highest average content) and shall be deemed to be more than 35 per cent. unless the contrary has been proved by an analysis made and communicated to the Inspector within the previous 12 months. The analysis shall be made by one of the methods specified by the *British Standards Institution*, and the sample of coal used for such analysis shall be taken either from a representative section of the seam or from a representative quantity of the run-of-mine coal from the seam. (*Note.*—The effect of the temporary amendment to Regulation 10 is to substitute 25 per cent. in this definition.)

The foregoing requirements do not apply to anthracite mines or to certain other mines in which specified conditions (described in detail) obtain. The incombustible dust used for the purpose of these Regulations shall be:—(a) of such fineness that of the dry dust which passes through a 60-mesh sieve not less than 50 per cent. by weight and, except with the permission in writing of the Board of Trade, not more than 75 per cent. by weight shall pass through a 240-mesh sieve; (b) of such character that it is readily dispersable into the air, when in use in places where it is not directly wetted by water from the strata, and does not cake, but is dispersed into the air when blown upon with the mouth or by a suitable appliance.

No dust shall be used for the purpose of complying with the Regulations of a kind which may be prohibited by the Board of Trade on the ground that it is injurious to the health of persons working in a mine.

Another Regulation (6) prescribes steps to be taken to see that the Regulation (3) relating to the prevention, suppression, collection and removal of coal dust and for treating with incombustible dust are complied with.

Under Regulation 10 these Regulations shall come into force on January 1st, 1940, *except that in so far as any provision requiring the dust to contain more than*

* Regulations, Mine Dust Analysis Order and Memorandum on Methods of Analysis. M. & Q., Form No. 128, December, 1939. London: H.M. Stationery Office. Price 6d. net.

65 per cent. of incombustible matter shall come into force on January 1st, 1941.
(Note.—The provision in italics is temporarily suspended.)

SCHEDULE (REGULATION 3)

MINIMUM PERCENTAGE OF INCOMBUSTIBLE MATTER REQUIRED FOR COALS OF VARIOUS VOLATILE MATTER CONTENT

| Average volatile matter content of coal Per Cent. | Minimum percentage of incombustible matter required |
|--|--|
| Not exceeding | |
| 20 | 50 |
| 22 | 55 |
| 25 | 60 |
| 27 | 65 |
| 30 | 68* |
| 32 | 70* |
| 35 | 72* |
| Exceeding | |
| 35 | 75* |

PART II. ORDER OF THE BOARD OF TRADE PRESCRIBING METHODS OF ANALYSIS OF MINE DUSTS.—The sample shall be sieved in a specified manner through a 60-mesh sieve, or, if too damp, through an 18-mesh sieve, and the fraction passing through shall be allowed to dry for an hour and thereafter sieved through a 60-mesh sieve; a correction is applied for the percentage loss of moisture.

(A) *Dust samples which contain no Carbonates or Gypsum.*—A weighed quantity is dried between 105° and 110° C. and the loss in weight is reckoned as moisture. The residue is heated at red-heat in an open vessel until it ceases to lose in weight. The weight of the incinerated residue added to the weight of moisture gives the amount of incombustible matter; it shall be expressed as a percentage of the total weight of the sieved dust.

(B) *Dust samples which contain Carbonates.*—(i) The moisture is determined at 105–110° C. (ii) The residue is heated at 950° C. (at least) until it no longer loses weight. (iii) The weight of carbon dioxide evolved from the dust is determined directly or calculated from the volume of carbon dioxide (or by other method approved by the Board of Trade). (iv) The sum of the weights of moisture, carbon dioxide and incinerated residue are reckoned as incombustible matter.

(C) *Dust samples which contain Gypsum.*—(i) The sample of sieved dust is dried between 135° and 140° C. and the weight lost is reckoned as moisture. (ii) The residue is heated at red-heat in an open vessel until it ceases to lose weight. The sum of the moisture and incinerated residue is reckoned as incombustible matter.

(D) *Dust samples which require special Methods of Analysis.*—These are to be analysed by such methods as shall be prescribed by the Board of Trade.

PART III. A DESCRIPTION OF APPROPRIATE ANALYTICAL METHODS.—*Moisture-Content.*—The methods of determining the moisture in coals by heating in nitrogen or in a partial vacuum (British Standard Specification, No. 735, of 1937), may be used for mine dusts. If, however, the dust contains gypsum the moisture must be expelled between 135° and 140° C. (or with advantage at 180° C. if air is rigorously excluded). The following method, however, is sufficiently accurate for the purpose:—One g. of the dust is spread over the bottom of a silica dish (2 inches in diameter) provided with a well-fitting cover. The dish without its cover is heated at 105° to 110° C. (or at 135° to 140° C. if the dust contains gypsum) in a suitable oven for 1 hour, after which it is covered, cooled in a desiccator and weighed.

* The effect of the temporary amendment to Regulation 10 is to substitute a figure of 65 per cent.

Ash-Content.—The dish containing the dried dust is heated without its cover in a muffle furnace at 900° to 950° C. (not less than 950° C. if carbonates are present) until constant in weight. Provision should be made for the free circulation of air over the dust. When the volatile matter has been expelled, the final incineration is completed in the hottest part of the muffle.

Carbon Dioxide.—Dusts containing carbonate are decomposed with acid in a calcimeter, and the volume of carbon dioxide is measured and corrected to normal temperature and pressure. A suitable form of calcimeter is described and illustrated, and a separate nomogram for converting the volume of carbon dioxide into its percentage in the dust is provided in the pamphlet.

The Mines Department will welcome any suggestions for additions or further explanations that would make the memorandum on analysis more useful.

British Non-Ferrous Metals Research Association

QUANTITATIVE SPECTROGRAPHIC ANALYSIS WITH THE MICROPHOTOMETER*

THE Association has published a booklet that is well worth studying by anyone using or contemplating the use of spectrographic methods for quantitative analysis. The use of a microphotometer is a refinement of spectrographic technique which is only justified when the many factors that may affect accuracy and reproducibility have been studied and are under control.

The subject matter of the report falls under three main headings. First, a review of published information is given in some detail, this being classified in short sections, each dealing with an aspect of spectrographic technique of importance in quantitative work. Attention cannot be drawn to all the fourteen sub-headings, but such matters as "size and shape of electrodes," "pre-sparking period," "calibration of the photographic plate," and "effects due to alloying constituents" will give some indication of the scope of the discussion.

Secondly, a summary of published procedures is given for a large number of alloys listed under the following principal constituents:—aluminium, copper, lead, magnesium, nickel, tin and zinc. The accuracy claimed for the method and a reference to the original work are given in each instance.

The third part consists of a bibliography of recent papers dealing with applications of the microphotometer. It is of considerable value to have references dealing with a specialised branch of one subject collected in this manner.

B. S. C.

* Part I. A Review of Published Work, by D. M. Smith, B.Sc., D.I.C., F.Inst.P. (British Non-Ferrous Metals Research Association; Research Report, Association Series, No. 524.)

Olive Oil Specifications for the Norwegian Canning Industry*

CANNING tests have shown that types of oil that are suitable for French and Portuguese sardines are not necessarily suitable for Norwegian brisling and sild. The Norwegian trade associations concerned with the canning of brisling and sild require all fish to be packed in olive oil answering to the following specifications:

- (1) The oil must be clear, free from water, pulp, or other impurities; *i.e.* it must be carefully filtered.
- (2) The colour must be pale yellow with only a tinge of green or brown.
- (3) Flavour and odour must be pure and good.
- (4) The acid-content (calculated as percentage of free fatty acid) must not exceed 1.7 per cent., assuming that the oil consists exclusively of expressed (not chemically refined) oil.
- (5) The content of chemically refined expressed oil used for blending must not exceed 30 per cent. of the mixture. The chemically refined oil must be of good quality.
- (6) The oil must not contain refined extracted oil (sulphur oil).
- (7) The oil must be resistant to cold; *i.e.* there must be no appreciable deposit of solid fat when the oil is cooled to between 4° and 8° C.
- (8) The iodine value must not exceed 88.
- (9) The rancidity must not exceed 10 red Lovibond units in the quantitative Kreis test.
- (10) The ash-content must not exceed 30 mg. per litre.

With regard to Specification 5, which, it is stated, is the one most disputed, it is pointed out that if chemically refined oil is used as the major part of the mixture, it may, unless it is of absolutely first-class quality, cause deterioration of the oil on storage. The limit of 30 per cent. as the maximum proportion in the mixture is in accordance with the views of well-known olive oil experts in Tortosa, Reus and Borjas Blancas. Moreover, systematic canning tests with brisling and sild have shown that, even if mixtures containing 80 per cent., or more, of refined expressed oil are used by Spanish and French canners, large quantities of the refined oil are not so suitable for the Norwegian fish.

The quantity of refined expressed oil in a mixture is estimated by an optical method (due to Lunde and Stiebel), and the results are expressed in terms of maximum blue fluorescence. If the figure is less than 130 the oil will be accepted as pure expressed oil. The figure increases with the amount of refined oil, up to about 600 for pure chemically refined expressed oil (*cf. Z. angew. Chem.*, 1933, **46**, 243, 796).

With regard to Specification 7, experience has shown that Spanish oils, whether from the north or south, usually show no tendency to deposit stearine under the conditions given. Oils from Tunis and Algeria (French oils) do not remain so clear as Spanish oils, and the quantity of stearine deposited during the test increases with the locality of the oil from north to south in those countries.

* Laid down by the Research Laboratory of the Norwegian Canning Industry. E. Mathiesen. *Tidsskrift for Hermetikindustri*, Jan., 1940, 10-12.

British Standards Institution

WE have been asked to give prominence to the particulars of the following Standard Specification:

BS/ARP 27. TESTING INCOMBUSTIBLE MATERIAL RESISTANT TO INCENDIARY BOMBS*

With a view to minimising the dangers resulting from possible incendiary bomb attacks of the enemy, active consideration has been given by the A.R.P. Department of the Ministry of Home Security to the development of materials which could be used in the attics and roof spaces of buildings so as to confine and afford protection against the incendiary effect of the bomb.

In order to assist in the development of such materials a test has been devised by means of which the performance of materials under incendiary bomb effect may be ascertained.

This method of test, together with the results which should be expected from a suitable material, has been made the subject of a standard in the BS/ARP series, which has just been issued by the British Standards Institution as BS/ARP 27.

It is stated in a note to the Standard that the method of test is one that has been in use for some time at the testing station of the Fire Offices' Committee, and it is indicated that the tests on materials will be made at the Elstree Testing Station on behalf of manufacturers who would like to have their materials examined.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Milk Fat from the Turkish Buffalo. A. Heiduschka and F. Cicekdagi. (*Z. Unters. Lebensm.*, 1940, 79, 150-153.)—Buffalo milk constitutes 8.3 per cent. of the total milk production of Turkey. In comparison with cows' milk it has a higher and more variable fat-content, ranging from 4 to 10 per cent. with an average of about 7.6 per cent. It is rarely used in the fresh state because its high fat-content renders it somewhat indigestible. The butter is usually prepared from Yoghurt and the butter-milk is converted into cheese. The milk is also used for the preparation of a very thick cream (Kaymak). The butter is pale, sometimes almost white, with a pleasant cream-like taste and a good aroma. The butter-fat gave the following constants:—sp.gr., 0.9273; m.p., 47.6° C.; solidifying p., 41.3° C.; n_D^{20} (Zeiss-Wollny), 41.3; saponification equiv., 318.6; iodine value (Hübl), 37.78; acetyl value, 32.33; Hehner value, 87; Reichert value, 31.5; Polenske value, 0.6; acid value, 3.8; total fatty acids, 94.05 per cent.; unsaponifiable matter, 0.165 per cent. The total fatty acids had the following composition (per cent.):—butyric acid, 4.24; caproic acid, 1.28; caprylic acid, 0.42; capric acid, trace; lauric acid, 2.99; myristic acid, 7.22; palmitic acid, 25.63; stearic acid, 16.18; arachidic acid, 3.24; oleic acid, 35.20; linolic acid, 1.98. This composition does not differ materially from that of the milk-fat of buffaloes in other districts.

A. O. J.

Ultra-violet Absorption of Yeast Extracts and Meat Extracts. J. Schormüller. (*Z. Unters. Lebensm.*, 1940, 79, 46-57.)—Numerous measurements of the ultra-violet absorption spectra of solutions of 5 yeast extracts from different sources gave curves with a maximum value at 260 to 262 $m\mu$ and a

* Copies of this BS/ARP Standard may be obtained from the British Standards Institution, 28, Victoria Street, London, S.W.1. Price 3d. post free.

minimum value at 233 to 236 $m\mu$. According to Heyroth and Loofbourow (*Nature*, 1934, 134, 461; *Biochem. J.*, 1936, 30, 65), vitamin B₁ exhibits 2 maxima, at 235 and 268 $m\mu$ respectively, and other authors have reported similar results. Quantitative considerations indicate that in the concentrations used (1 g. of extract in 1000 ml. of water) the amounts of the vitamins B₁ and B₂ are too small to account for the measured absorption. According to Williams (*J. Amer. Chem. Soc.*, 1935, 57, 1093) and Smakula (*Z. physiol. Chem.*, 1935, 230, 231) the maximum value at 260 $m\mu$ is characteristic of the pyrimidine ring of aneurin, and it is now suggested that since yeast extracts exhibit only one maximum value, pyrimidine bodies are principally responsible for the form of the curve. Such bodies are formed by decomposition of the nucleic acids of yeast during preparation of the concentrate. The ultra-violet spectrum of yeast nucleic acid showed remarkable resemblance to that of the extract. Aneurin alone is not therefore responsible for the absorption, and measurement of the maximum absorption is no indication of the vitamin B₁ content of the extract. A comparison of the curves given by yeast nucleic acid with those of the concentrates indicates that the extracts are much richer in pyrimidine bodies. Yeast gum was also investigated, but showed no characteristic absorption. The extract was separated by ultra-filtration into constituents of high and low molecular weight, and it was found that the characteristic portion of the curve is due to the low molecular constituents which include the decomposition products of the nucleic acids. Similar experiments with fractions separated by dialysis were less successful, but tended to confirm this conclusion. Heating in a boiling water-bath for 5 hours caused no change in the absorption curve of yeast extract. The absorption curves of meat extracts were compared with those of yeast extracts. Although the general course of the curves is the same, they do not exhibit distinct maximum or minimum values, but an inflexion-like bending of the curves suggests that the selective absorption of such decomposition products as tyrosine, tryptophane and phenyl-alanine is masked by other substances. By a study of the absorption curves of different mixtures of yeast extract and meat extract it was found that the characteristic form of the yeast extract curve was perceptible when 33 per cent. was present in meat extract, and as this percentage was increased the curve conformed more and more closely to that of pure yeast extract. Examination of the absorption curves of lactoflavin concentrates prepared from meat extracts showed that the absorption is masked by that of other substances, and spectrographic identification of vitamin B₂ in meat extract is thus not directly possible. The spectrographic method cannot therefore replace chemical and biological methods of determining vitamin contents, but can be used to distinguish the two types of extract and to detect admixture of yeast extract with meat extract.

A. O. J.

Presence of *l*-Tyrosine in the Alcoholic Extract of Egg Yolk.
L. Bracaloni. (*J. Pharm. Chim.*, 1940, 132, 140-142.)—Serum decanted from the paste of lecithin and lutein left after distillation *in vacuo* of an alcoholic extract of egg yolk contains a small amount of a nitrogenous substance which separates in white flakes (0.10 to 0.16 g. per 200 g. of egg yolk). Its micro-crystalline form, colour reactions (xanthoproteic reaction and reaction with

Millon's reagent), m.p., and percentage of nitrogen (by Kjeldahl's method) are those of *l*-tyrosine and with formic acid it forms a derivative resembling Fischer's formyl-*l*-tyrosine (*Ber.*, 1907, 40, 3716). Its elementary percentage composition also agrees with that of *l*-tyrosine. E. B. D.

Iodine Absorption of Honey and Artificial Honey. C. Griebel and G. Hess. (*Z. Unters. Lebensm.*, 1940, 79, 171-177.)—Only a few kinds of honey are able to decolorise considerable amounts of *N*/100 iodine solution. Thyme honey and mint honey show pronounced absorption and a weaker absorption is shown by honey dew, heather honey and buckwheat honey. If the iodine absorption is compared with the titration with dichlorophenol-indophenol it is found that with mint and thyme honeys the ascorbic acid titre is only slightly lower than the iodine absorption, but with honey dew, buckwheat and heather honeys it is considerably lower. Iodine titration yields results agreeing closely with the dichlorophenol-indophenol titration if a relatively large amount of potassium iodide is added to the honey solution before titration. The iodine absorption figure obtained in the absence of potassium iodide is not independent of the amount of sample used, and it is therefore necessary to work with the same amount of sample, e.g. 5 g., except with mint and thyme honeys where 1 to 2 g. is sufficient. The difference between the iodine absorption with and without the addition of potassium iodide is considerable for most honeys, but the cause of the difference is not yet known. An investigation was made in the following manner:—The honey (5 g.) was dissolved in 25 g. of cold 2 per cent. metaphosphoric acid, which served to precipitate albuminous substances and to stabilise the ascorbic acid. The solution was titrated with *N*/100 iodine solution in presence of starch, and when the end-point was reached 1 ml. of iodine solution was added in excess and, after the lapse of 30 sec., the mixture was titrated back with *N*/100 sodium thiosulphate solution. The result of this titration calculated for 100 g. of honey was denoted by *a*. The experiment was then repeated with the difference that 1 g. of potassium iodide was added before titration. The corresponding value found by this titration was denoted by *b*. With buckwheat honey the metaphosphoric acid precipitated a considerable amount of albuminous matter. This was separated by centrifuging, and weighed aliquot portions of the centrifugate were taken for titration. With artificial honey the value found for *b* was nil and the value of *a*-*b* rarely exceeded 5. With native honey (excluding heather, buckwheat and the labiate honeys and honey dew) the value of *b* varied from 1 to 4.8, and the value of *a*-*b* was, as a rule, greater than 5, but rarely exceeded 10. Among foreign honeys two samples (Canada White Clover 1939 and Salvador 1938) gave no absorption after the addition of potassium iodide. The value of *b* varied from 0 to 4, and the highest value of *a*-*b* was 9. With heather honey *b* always exceeded 5, and *a*-*b* ranged from 4 to 9. With honey dew (excluding some samples with an odour of floral honey) the value of *b* varied from 5 to 16, and that of *a*-*b* from 9 to 20. With buckwheat honey *b* varied with the source from 5.8 to 16, and *a*-*b* from 8 to 48. The value of *a*-*b* was exceptionally high for Canadian and Hungarian buckwheat honey. Since such large values do not occur with other honeys, it may be assumed that a value for *a*-*b* of over 30 indicates buckwheat honey. With buckwheat

honey it was found that the value of b rose after 4 months' storage, and that the value of a diminished, so that the value of $a-b$ also diminished considerably. The following conclusions may be drawn:—Iodine absorption after the addition of potassium iodide is not influenced by invert sugar because most artificial honeys give no absorption. Although the value of b for artificial honey is either nil or very low, it is possible to conclude that artificial honey is present in a sample only if the value of $a-b$ exceeds 5, because low absorption and difference-values up to 5 occur with artificial honey owing to the presence of added natural honey. Since only a few kinds of honey (heather and buckwheat honeys and honey dew) show a greater iodine absorption in presence of potassium iodide than the floral honeys, and since buckwheat and honey dew give difference-values that generally exceed 10, these may be recognised by their behaviour towards iodine. Verification by pollen analysis is necessary, however, because raising of the absorption value may be due to the addition of labiate honey.

A. O. J.

Detection of Artificial Dyes in Tomato Pulp. H. Thaler and K. E. Schulte. (*Z. Unters. Lebensm.*, 1940, **79**, 74–77.)—Since tomato pulp is manufactured from unripe or imperfect fruit, the colour is frequently intensified by the addition of water-soluble artificial dyes. In order to determine whether such addition can be detected by adsorption analysis, preliminary experiments were made with the natural pigment of the tomato. Aluminium oxide (Merck's technically pure hydroxide heated in a shallow dish at about 400° C. until no more steam was evolved) in a column, 20 cm. high and 1 cm. broad, was used as the adsorbing substance, and the solvents employed were pyridine, benzene, carbon disulphide, ether, petroleum spirit, methyl alcohol, 70 per cent. ethyl alcohol and acetone. The artificial dyes used in the investigation were Tomato Red (3242) and Lobster Red (*Krebsrot* 2872). These were insoluble in benzene, ether, carbon disulphide and petroleum spirit, but soluble in pyridine, methyl alcohol, 70 per cent. ethyl alcohol and acetone. It was found that methyl alcohol, ethyl alcohol and acetone gave the best results, because the natural pigment when dissolved in these is not characteristically adsorbed by the aluminium oxide, whilst the artificial dyes give characteristic chromatographs. For the colouring of the pulp a little of the dye was dissolved in water and incorporated with the pulp. After an hour a portion of the pulp was warmed with the solvent at 30° C. for half-an-hour and the extract was filtered. As a rule the artificial dyes imparted a strong red colour to the surface of the aluminium oxide and to the layers immediately beneath the surface. When Tomato Red was adsorbed from solutions in ethyl alcohol or acetone a yellow zone appeared beneath the red zones. If it is desired to verify that a preparation has been manufactured from undyed tomato pulp the solvents recommended are benzene and ether. From solution in benzene the natural pigment is adsorbed by the aluminium oxide and colours its surface yellow, and below are formed in order white, rose-coloured and red zones. With an ethereal solution of the natural pigment the order of colours is yellow, white, brownish-yellow, rose. The method was used successfully to prove that the colouring matter in a preparation of herrings in tomato was the natural pigment of the fruit.

A. O. J.

Thermo-stability of Fats. E. Glimm, H. Wittmeyer and W. Jahn-Held. (*Z. Unters. Lebensm.*, 1939, **78**, 285–293.)—The thermal decomposition of beef-fat, lard, coconut oil, palm oil, sesame oil, arachis oil, olive oil and triolein was studied at temperatures from 60° to 120° C., during periods of 7 days. It was found that fat-splitting by heat is independent of the percentage of free acids initially present. At temperatures below 60° C., no fat-splitting takes place, except in beef-fat and lard. The vegetable fats each show a definite decomposition temperature, at which after 3 days a pronounced increase in the acid value occurs. These temperatures are as follows:—for coconut oil, 100° C.; palm oil, 90° C.; olive oil, 90° C.; arachis oil, 80° C.; soya-bean oil, 80° C.; sesame oil, 75° C. Beef fat and triolein are remarkable in that the isotherms of acid-splitting in relation to time for beef-fat coincide for 60°, 75° and 90° C., and for triolein for 75°, 90° and 105° C. It seems that the splitting at certain temperature intervals starts with a jump. The results of the Kreis test indicate that at the beginning of the decomposition the proportion of aldehyde increases, but decreases on further decomposition. The amount of volatile fatty acids increases with rise of temperature, whilst that of unsaturated fatty acids decreases. Carbon dioxide has no influence on the thermal decomposition of lard, but has a retarding effect on the decomposition of vegetable fats.

D. A.

Action of a Cuprous Iodide Reagent on Alkaloids: Precipitation and Colour Reactions. M. Péronnet and J. Guénin. (*J. Pharm. Chim.*, 1940, **132**, 142–147.)—A cuprous iodide reagent for the detection of "yperite" ($\beta\beta'$ dichloro-diethyl sulphide) has been prepared as follows (*cf.* Grignard, Rivat and Scatchard, *Ann. Chim.*, 1921, **15**, 14): To 50 g. of a 30 per cent. solution of crystallised sodium iodide, 30 drops (about 1.23 g.) of a 7.5 per cent. solution of crystalline copper sulphate are added, with shaking. If the resulting solution becomes turbid after a few hours it is filtered; it will then keep indefinitely in the dark. With yperite this gives a yellowish-white precipitate of $\beta\beta'$ diiodo-diethyl sulphide. It has been found that the reaction is characteristic of yperite only if the diiodo compound is identified (small colourless prisms, m.p. 62° C.); the reagent forms insoluble compounds with most alkaloids, the precipitates being yellowish-white, yellowish or brown. The sensitivity values for various alkaloids are given. They vary from 1 in 500 (morphine hydrochloride) to 1 in 200,000 (sparteine sulphate). The principal glycosides and barbiturates do not form precipitates either in aqueous or in hydrochloric acid solution. Characteristic colour reactions are given by: (a) eserine and (b) ephedrine. If the precipitate that the cuprous iodide reagent forms with eserine hydrochloride solutions is dissolved in ammonia, there is produced a violet-red colour which changes slowly to brown. Sensitivity: 1 in 10,000. If the cuprous iodide reagent is added, drop by drop, to an aqueous solution of ephedrine, the reagent is first decolorised and then assumes a characteristic violet colour, which is very stable in neutral solutions but disappears on acidifying. Sensitivity: 1 in 10,000. Under the same conditions adrenaline gives a reddish colour which disappears on acidifying. For all the tests, 1 ml. of reagent and 4 ml. of the alkaloid solution (1 per cent., or more dilute according to solubility) were used.

E. B. D.

Study of *Centaurea scabiosa*. C. Charaux and J. Rabaté. (*J. Pharm. Chim.*, 1940, 132, 155-162.)—The fresh leaves of *Centaurea scabiosa* L. contain about 2 per cent. of a glycuronoside, $C_{21}H_{18}O_{12} \cdot 2H_2O$ (present as a water-soluble salt of an alkali or alkaline earth metal). Hydrolysis of this glycuronoside yields glycuronic acid and a flavonol identical with the scutellareol obtained by hydrolysis of scutellaroside, the glycuronoside of *Scutellaria altissima*. The physical properties of scutellaroside given by Goldschmiedt and Zerner (*Monatsh. Chem.*, 1910, 31, 439) differ from those of the centaury glycuronoside, but a scutellaroside prepared by the authors by the Goldschmiedt method and purified by crystallisation from 50 parts of boiling absolute alcohol was identical with the new product. The physical properties now found are:—rotation, $[\alpha]_D^{15}$, -128° ; $[\alpha]_D^{17}$, -129.5° (anhydrous, $[\alpha]_D^{15}$, -138°); m.p. 230° C. (Maquenne block). E. B. D.

Biochemical

Excretion of Volatile Selenium Compounds after Administration of Sodium Selenite to White Rats. J. Schultz and H. B. Lewis. (*J. Biol. Chem.*, 1940, 133, 199-207.)—It has frequently been asserted that, after administration of selenium salts, volatile methyl compounds are excreted through the lungs. This has now been confirmed experimentally on rats injected subcutaneously with a solution of sodium selenite at a level of 2.5 to 3.5 mg. per kg. of body-weight. After the injections the animals were placed in cages inside a respiration chamber, the gases from which were passed successively through two tubes containing hydrochloric acid (1:1), a tube containing 20 per cent. sodium hydroxide solution, and a large absorption tube containing glass wool and calcium chloride. The dry gases then entered a specially designed all-glass absorption tube containing 45 ml. of conc. sulphuric acid, and passed through a second tube containing sulphuric acid, which was connected with a vacuum water-pump. With the current of air properly regulated, all the selenium was found in the first absorption tube containing sulphuric acid. At the end of each experiment the acid was transferred to a Kjeldahl flask, 1 ml. of 20 per cent. hydrogen peroxide was added, and the mixture was gently heated for 20 to 30 minutes. After cooling, the volume of the digest was measured, and 10-ml. portions were transferred to test-tubes. Two drops of an aqueous 4 per cent. solution of codeine phosphate were added, and the solution was cooled under the tap. The tubes were stoppered and placed in the dark for 20 to 30 minutes. The colours were compared in a colorimeter with that developed by 10 ml. of a standard solution of sodium selenite in conc. sulphuric acid. When known concentrations of sodium selenite were tested, discrepancies up to 15 per cent. were found between the observed and the theoretical values, but the method was sufficiently accurate for the investigation in hand. The results showed that 17 to 52 per cent. of the selenium injected was excreted within 8 hours in the form of a volatile selenium compound that could be absorbed by conc. sulphuric acid. With two possible exceptions, the simultaneous administration of either methionine or choline chloride failed to influence the excretion. The nature of the volatile selenium compounds remains to be elucidated. F. A. R.

Basic Amino Acid Content of Human Serum Proteins. Influence of the Ingestion of Arginine on the Composition of the Serum Proteins. R. J. Block. (*J. Biol. Chem.*, 1940, 133, 71-74.)—Dirr (*Z. physiol. Chem.*, 1939, 260, 65) reported that the intravenous or oral administration of arginine hydrochloride resulted in a considerable increase in the arginine content of the serum proteins. This observation could not be confirmed by the author. Samples of human blood were drawn before and after administration of arginine hydrochloride, and the serum was removed by centrifuging after the blood had clotted. The sera were acidified with 5 *N* acetic acid to *pH* 4.5 and, after the addition of 3 volumes of dilute sodium chloride solution, the proteins were coagulated by heat. The precipitate was washed 3 times with hot water, and the lipids were removed by extraction with acetone, hot alcohol, hot benzene, and ether. The basic amino acids were determined by the author's silver precipitation method ("*The Determination of the Amino Acids*," Minneapolis, 1938). Arginine was isolated as the flavianate, histidine as the nitranilate, and lysine as the picrate. The molecular ratios of histidine to arginine to lysine were unaffected by the feeding of arginine. The same result was obtained when the proteins were isolated by precipitation with alcohol, instead of by heat coagulation. F. A. R.

Activation of Papain. J. S. Fruton and M. Bergmann. (*J. Biol. Chem.*, 1940, 133, 153-156.)—The experiments described throw considerable doubt on the oxidation-reduction theory of papain activation. An inactive papain preparation (Papain A) was activated with hydrogen cyanide (HCN—Papain A) and subsequently precipitated with isopropyl alcohol (Papain B). If the activation of papain with hydrogen cyanide consisted merely in the reduction of disulphide groups to sulphhydryl groups, then Papain B should have been as active as HCN—Papain A. It scarcely showed any activity at all, however, but regained nearly all the activity of the original HCN—Papain A on addition of hydrogen cyanide. This activated Papain B was precipitated by isopropyl alcohol, and the precipitate (Papain C) was again inactive, but capable of being activated with hydrogen cyanide to a product with almost the same activity as the original HCN—Papain A. Cysteine was also used as activator, with similar results. The authors suggest that the precipitation with isopropyl alcohol brings about dissociation of the active HCN—enzyme compound with regeneration of the original inactive enzyme. It seems probable that the specificities of the various activator-enzyme compounds differ, depending on the nature of the activator. The results of these experiments support the theory of Mendel and Blood (*J. Biol. Chem.*, 1910-11, 8, 177), that hydrogen cyanide and other activators serve as co-enzymes for papain. F. A. R.

Estimation of Vitamin A and Carotene with the Photoelectric Colorimeter. C. J. Koehn and W. C. Sherman. (*J. Biol. Chem.*, 1940, 132, 527-538.)—Dann and Evelyn (*Biochem. J.*, 1938, 32, 1008; *cf.* ANALYST, 1938, 63, 611) first used the photoelectric colorimeter to measure the intensity of the blue colour obtained by the action of antimony trichloride solution on vitamin A, obtaining a factor of 0.41 ± 0.05 for the conversion of $L_{1\text{cm}}^1$ (620 $m\mu$) into $E_{1\text{cm}}^1$ (328 $m\mu$). In view of the controversy over the value of the factor required to convert $E_{1\text{cm}}^1$.

(328 $m\mu$) into biological units, it was decided to determine directly the relationship between $L_1^{1\% \text{ cm.}} (620m\mu)$ and biological activity, and for this purpose $L_1^{1\% \text{ cm.}} (620m\mu)$ of the U.S.P. reference cod-liver oil was determined and its potency checked by biological assay against β -carotene. At the same time the $L_1^{1\% \text{ cm.}} (440m\mu)$ of β -carotene was determined in chloroform and "Skellysolve" solutions, and a correction factor was also determined for the amount of light absorbed by the blue colour produced by the reaction of carotene with antimony trichloride solution. The U.S.P. reference oil was found to have a biological potency of 3000 I.U. of vitamin per g., and its $L_1^{1\% \text{ cm.}} (620m\mu)$ was 3.45, whence, using the factor 0.41 for converting $L_1^{1\% \text{ cm.}} (620m\mu)$ into $E_1^{1\% \text{ cm.}} (328m\mu)$, a factor of 2120 is obtained for converting $E_1^{1\% \text{ cm.}} (328m\mu)$ into I.U. of vitamin A per g. This is in close agreement with the value of 2150 found by Mead, Underhill and Coward (*Biochem. J.*, 1939, 33, 589) as a result of work with vitamin A-2-naphthoate. β -Carotene was found to have an average $L_1^{1\% \text{ cm.}} (440m\mu)$ of 1645 and 1980 in chloroform and "Skellysolve" respectively. It was found that $2 - \log G$ (G is the corrected galvanometer reading) was not a strictly linear function of the concentration of vitamin A or carotene, but constants were found for use within a certain range of galvanometer readings, which do not incur an error of more than 4 per cent. For more accurate work the use of a calibration curve is recommended.

F. A. R.

Ascorbic Acid Content of Rose Hips. W. Goldberg and E. O'F. Walsh. (*Pharm. J.*, 1938, 551.)—Ascorbic acid estimations were carried out on three different samples of rose hips by the 2:6-dichlorophenol-indophenol method, the procedure described by M. Olliver (*ANALYST*, 1938, 63, 2) being followed. The first sample, consisting of fruits that were not quite ripe, was collected early in September and was not limited to the fruit of any particular species of rose. The second sample was collected a fortnight later and consisted of ripe fruits of a similar character. The third sample was purchased, and on subsequent enquiry proved to consist of kiln-dried hips from Germany. The unripe and ripe freshly-collected hips contained respectively 385 and 364 mg. of ascorbic acid per 100 g., whereas the kiln-dried fruits contained only 64 mg. per 100 g. The fresh material lost 80 to 90 per cent. of its vitamin content on drying at 40° C., but only about 50 per cent. on drying at 60° C. The seeds contained no ascorbic acid.

F. A. R.

Estimation of Adermin (Vitamin B₆) in Urine. J. V. Scudi, H. F. Koones and J. C. Keresztesy. (*Proc. Soc. Exp. Biol. Med.*, 1940, 43, 118–122.)—In an investigation designed to study the excretion of adermin in the rat, it was necessary to find a method of assaying the vitamin. This was successfully achieved by treatment with 2:6-dichloro-quinone chlorimide reagent, and measurement of the resulting colour in a photoelectric colorimeter. The urine samples were made strongly alkaline to thymol blue (pH above 9.6) with 30 per cent. sodium hydroxide solution. After being allowed to stand overnight, 1-ml. aliquot portions of the urine were neutralised to pH 7 to 7.5, and the volume was adjusted to 25, 50 or 100 ml. as necessary. For the adjustment of the pH , bromothymol blue was used as external indicator. To 5 ml. of the diluted urine were added 5 ml. of veronal buffer (prepared by dissolving 18 g. of sodium diethyl barbiturate in 700 ml.

of water and titrating to pH 7.6 with dilute hydrochloric acid using the glass electrode, the solution being filtered from the precipitated barbituric acid) and 20 ml. of a butyl alcoholic solution of the chlorimide reagent (100 mg. of 2:6-dichloroquinone chlorimide dissolved in 1600 ml. of acid-free butyl alcohol). The tubes were briefly but vigorously shaken, and after 5 minutes were shaken again. After an additional 10 minutes the two layers were separated by centrifuging and the supernatant butyl alcohol layer was pipetted into 10 ml. of a fresh veronal solution. After shaking, the two layers were again separated by centrifuging, and the washing process was repeated. Fifteen ml. of the washed butyl alcohol layer were pipetted into a colorimeter tube containing 5 ml. of absolute ethyl alcohol, the contents were thoroughly mixed and the colours were measured 40 minutes after the addition of the reagent in an Evelyn colorimeter, filter No. 660 being used. Under these conditions 100 per cent. recoveries were obtained when the pure vitamin was added to urine.

F. A. R.

Vitamin K Activity of 4-Amino-2-Methyl-1-Naphthol and 4-Amino-3-Methyl-1-Naphthol. A. D. Emmett, O. Kamm and E. A. Sharp. (*J. Biol. Chem.*, 1940, 133, 285-286.)—The hydrochlorides of 4-amino-2-methyl-1-naphthol and 4-amino-3-methyl-1-naphthol are readily soluble in water or saline solution, and can be administered either orally or parenterally. The former was found to have a potency of 1200 units per mg., expressed in terms of 2-methyl-1,4-naphthoquinone as standard (1000 units per mg.), and the latter a potency of 780 units per mg. Thus the former is about 3 times as active as vitamin K₁; it has given excellent results clinically, both in obstructive jaundice and in neonatal haemorrhage. [Dam, Glavind and Karrer (*Helv. Chim. Acta*, 1940, 23, 224), on the other hand, report methyl-naphthoquinone to be 2½ times as active as the amino-compound.—*Abstractor.*]

F. A. R.

Agricultural

Determination of Iodine in Soils, Plant Material and Waters. G. S. Fraps and J. F. Fudge. (*J. Assoc. Off. Agric. Chem.*, 1940, 23, 164-171.)—Objections to the A.O.A.C. tentative methods for the determination of iodine in soils, plant materials and brine (*Methods of Analysis*, A.O.A.C., 1935, 8, 133, 528) arise mainly from losses of iodine or from interference by other substances with the liberation of iodine (*cf.* ANALYST, 1935, 60, 631). The following method has therefore been adopted by the Texas Agricultural Experimental Station, and has the additional advantage of ease of manipulation. The organic matter is removed by burning and wet oxidation, and the iodine, which is thereby oxidised to iodic acid, is subsequently reduced with phosphorous acid and separated by distillation; it may then be determined colorimetrically. Plant materials are first burned (*cf.* Von Kolnitz and Remington, *Ind. Eng. Chem., Anal. Ed.*, 1933, 5, 38), and the solution, containing the ash and washings, is evaporated with sodium hydroxide to about 30 ml. Waters (500 ml.) are evaporated directly with the alkali, and soils are diluted with 30 ml. of water. To the resulting mixture in each instance are added 6 g. of potassium chromate, 30 ml. of a solution of 3 g. of potassium chromate in a mixture of 95 ml. of conc. sulphuric acid and 5 ml. of water, and

10 mg. of cerous sulphate which has been washed 3 times with purified alcohol. The mixture is maintained at 195° C., with occasional shaking, and when all the organic matter has been destroyed (after about 1 hour), 50 ml. of water are added and the precipitate is removed by filtration through asbestos which has undergone the same digestion process. The filtrate is cooled to below 50° C., and transferred to an all-glass distillation apparatus (see below), and sufficient phosphorous acid (about 10 ml.) is added to destroy the yellow colour. The solution is heated, air being drawn through it by suction, slowly at first and then, as distillation proceeds, at the rate of 50 to 75 litres per hour, so that the solution is violently agitated. The receiver contains 10 ml. of 0.02 *N* sodium hydroxide solution, and distillation should be stopped after 30 minutes, when the temperature of the solution should have reached 150° C. The distillate is neutralised with 50 per cent. sulphuric acid, one drop being added in excess, followed by sufficient bromine to produce a brown colour. The solution is then evaporated to 5 ml., and 5 ml. of water, 4 ml. of the dilute sulphuric acid and 1 ml. of 0.1 per cent. potassium iodide solution are added. This mixture is shaken vigorously with 1 ml. of carbon tetrachloride for 2 minutes, and the extract is separated and centrifuged for 1 minute. The colour may then be matched against a series of standards which are prepared in exactly the same way from known amounts of iodine. It is advisable to purify the carbon tetrachloride by adding 50 ml. of bromine water to 500 ml., and allowing the mixture to stand for several hours in sunlight; it is then made alkaline with sodium hydroxide, and on the following day is washed 3 times with water and dried by shaking with plaster of Paris, and the decanted solvent is distilled, the first and last 25 ml. being rejected. The distillation apparatus (*cf.* Trevorrow and Fashena, *ANALYST*, 1935, 60, 628; *J. Biol. Chem.*, 1936, 114, 351) consists of a 150-ml. round-bottomed flask, which is connected with a train consisting of (1) an inverted 250-ml. Erlenmeyer flask, into the base of which is sealed (2) a bulb, into which is sealed a Kjeldahl connecting tube, and (3) a condenser, which is connected by means of a ground-glass joint with (4), the cylindrical receiver (capacity, 100 ml.). The technique of the individual steps of the reaction is fully discussed. Comparison with the McHargue method (in which the iodine is removed from the soil by volatilisation in an electric furnace; *cf.* *J. Assoc. Off. Agric. Chem.*, 1937, 20, 222) for 3 sandy loams containing 1.1 to 4.5 p.p.m. and one black clay containing 13.4 p.p.m. of iodine, showed that the present method gave results that were higher by 0.1 to 0.6 and 0.7 p.p.m., respectively. In another test different quantities of the same samples of water were used, *viz.* aliquot portions of 500 and 2000 ml. The average iodine contents of 8 samples containing 4 to 25 parts per 10⁹ were 15.6 and 16.4 p.p.10⁹ for the respective aliquot portions; the greatest difference (4 p.p.10⁹) was obtained with a sample containing 20 p.p.10⁹ of iodine.

J. G.

Organic

Qualitative Test for Organic Compounds containing Oxygen.

D. Davidson. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 40-41.)—Ferric thiocyanate dissolves in oxygen compounds, yielding a red solution, but is insoluble in hydrocarbons and organic halogen compounds. *Method.*—Filter-paper is saturated

with a solution of 1 g. of ferric chloride and 1 g. of potassium thiocyanate in 10 ml. of methanol (which has been filtered to remove precipitated potassium chloride) and air-dried. More than one dipping may be necessary to produce a paper having a greenish hue resembling fuchsine crystals. The test-paper is kept in a stoppered bottle away from sunlight. A small piece of the paper is stirred with a few drops of the liquid to be tested. Solids are tested as saturated solutions in a hydrocarbon or halogenated derivative. The production of a dark red solution indicates the presence of an oxygen compound, provided that nitrogen and sulphur compounds are absent. Nitrogen compounds, such as the amines, also dissolve the reagent. Few sulphur compounds have been tested, but among these carbon disulphide reacted negatively, whilst benzyl sulphide gave a positive reaction. The name "ferrox" is proposed for the method. S. G. C

Detection of Elements in Organic Compounds. L. Rosenthaler. (*Pharm. Acta Helv.*, 1939, 14, 215-216; *J. Pharm. Belg.*, 1940, 22, 220.)—**Bromine and Chlorine.**—The test substance is mixed with several drops of a saturated solution of potassium permanganate and 1 ml. of conc. sulphuric acid; chlorine or bromine is liberated. The bromine may be identified by means of fluorescein paper (formation of eosin and subsequent bleaching) which is merely bleached by chlorine. Chlorine gives characteristic crystals when it acts upon a 1 per cent. solution of 1, 2, 5-toluylene diamine in dilute sulphuric acid on a microscope slide; bromine does not give this reaction. The bromine test is successful with most bromine compounds except tertiary butyl bromide; exceptions for chlorine include: carbon tetrachloride, *p*-chlorophenol, chloro-*m*-cresol and *p*-chlorobenzoic acid. **Sulphur.**—The action of nascent hydrogen, with formation of hydrogen sulphide (detected by its action on lead acetate paper), is not a characteristic reaction for sulphur in organic compounds. The method of conversion into thiocyanate is being investigated. (*Abstractor's note.*—For satisfactory test for sulphur see Feigl, "*Spot Tests*," 2nd Eng. Edn., 1939, p. 262.) **Carbon.**—The usual test is charring with sulphuric acid. The following substances do not give this reaction:—urea, thiourea, hydrocyanic acid, dimethylglyoxime, diphenylurea, benzene, sulphobenzoic acid, phenol, salicylic acid, sulphosalicylic acid, *p*-hydroxybenzoic acid, anisic acid, coumarin, phthalic anhydride, hydroxyquinoline and acridine. Ethyl alcohol is more readily attacked (150° C.) than methyl alcohol (230° C.). J. W. M.

Preparation, Properties and Thiocyanogen Absorption of Triolein and Trillinolin. D. H. Wheeler, R. W. Riemenschneider and C. E. Sando. (*J. Biol. Chem.*, 1940, 132, 687-699.)—The triglyceride of oleic acid was prepared by direct esterification of oleic acid and glycerol in an atmosphere of nitrogen with *p*-toluene sulphonic acid as catalyst. It was purified by molecular distillation, the fraction with the theoretical iodine number (86.1) having the following properties: saponification equiv. 295.8 (theory 294.9); n_D^{40} , 1.4621; n_D^{60} , 1.4586; density at 40° C., 0.8988. From a study of the melting- and freezing-points of triolein, it was observed that polymorphism existed, there being apparently three crystalline or solid forms: I, the stable modification, m.p. 4.7 to 5.0° C.; II, m.p. about -12° C.; III, m.p. about -32° C.

The triglyceride of linolic acid was also synthesised and purified in the same way. It had the following properties: saponification equiv. 292.4 (theory 292.9), iodine value 173.9 (theory 173.4), n_D^{40} 1.4719, n_D^{50} 1.4683, density at 40° C. 0.9184. Two solid forms were observed: I, m.p. -13.1 to -12.8° C., and II, m.p. about -43° C. Thiocyanogen values were determined by the modified Kaufmann method (*J. Assoc. Off. Agric. Chem.*, 1938, **21**, 87). It was found that a 4-hour reaction time is most suitable for both glycerides, the value obtained for triolein (85.8) being slightly low (theory 86.1) and that for trilinolein (87.1) slightly high (theory 86.7). Bromination of trilinolin in ethereal solution produced a number of bromine addition products, a 9.1 per cent. yield of crystalline bromides being obtained. These had m.p. 81.0 to 81.7° C.

F. A. R.

Separation and Determination of 4-Aminodiphenylamine. I. S. Shupe. (*J. Assoc. Off. Agric. Chem.*, 1940, **23**, 161-164.)—The monohydrochloride of 4-aminodiphenylamine, which is a common constituent of certain hair dyes, forms a dark blue solution on oxidation. The free base is relatively insoluble in water, very soluble in chloroform, ether or benzene, and slightly soluble in petroleum spirit, which can therefore be used for its isolation, as follows:—A solution of a quantity of the sample (equivalent to not more than 0.1 g. of the free base) in warm water or dilute hydrochloric acid is made alkaline with an excess of powdered sodium bicarbonate, and extracted with one 50-ml. and four 20-ml. portions of petroleum spirit. The combined extracts are washed with 20 ml. of water, and the aqueous-extract is washed once with 20 ml. of petroleum spirit, which is added to the other petroleum spirit extracts. The united extracts are then filtered through cotton-wool into a tared dish and evaporated on a water-bath in a stream of carbon dioxide, and the residual 4-aminodiphenylamine is dried at 100° C. for 30 minutes and weighed. It has a characteristic needle-shaped structure, and its m.p. is 66° to 67° C. if it is not dried at 100° C., and 73° to 74° C. if dried in this way and then recrystallised, or if it is distilled in a vacuum at a low temperature. These differences correspond with isomeric modifications of the base, the modification of higher m.p. being also obtained if an extract of the base in chloroform is sublimed in a vacuum. Both isomers give the same acetyl and benzene-sulphonyl derivatives, which are obtained as follows:—To a solution of a weighed portion of the extracted base in 5 ml. of 1 per cent. hydrochloric acid are added 0.5 ml. of acetic anhydride for every 0.1 g. of base, and then 1 g. of sodium bicarbonate. The excess of acetic anhydride is removed on the water-bath, 5 ml. of water are added to the residue, and after 1 hour in the cold the mixture is filtered on a tared Gooch crucible, 25 ml. of water being used for transferring and washing the acetyl derivative, which is then dried at 100° C. for 1 hour and weighed. It is slightly soluble in water, and, if recrystallised from a mixture of alcohol and water, has m.p. 162° to 163° C. (*cf.* Deshusses, below). If 0.7 mg. is added to the weight of derivative, the factor 0.8142 may be used to calculate the equivalent of free base. The benzene-sulphonyl derivative is obtained by adding to a solution of the base in 5 ml. of alcohol, 0.5 ml. of benzene sulphonyl chloride for every 0.1 g. of base, and 5 ml. of 25 per cent. sodium acetate solution. The mixture is heated on the water-bath for 30 minutes; 30 ml. of water are then added, and the

mixture is stirred at intervals during 1 hour. The precipitated derivative (m.p. 138°–139° C.) is separated, weighed and recrystallised as described above; the factor 0.5679 gives the free base (no correction required). The recovery obtained from 99.2 mg. of the pure salt was 99.3 per cent.; with a mixture of 49.6 mg. of the salt with 300 mg. each of *m*- and *p*-phenylene diamines and 2.5-diaminotoluene, 60 mg. each of *o*- and *p*-aminophenols, 30 mg. of 2,4-diaminoanisole and 100 mg. of *p*-methylaminophenol the recovery was 99.8, or 103.8 per cent. if sodium hydroxide was used to make the solution alkaline before extraction. With chloroform as solvent the recovery from 116.0 mg. of the pure salt was 100.0 per cent. Recoveries based on the weights of the acetyl and benzenesulphonyl derivatives were 99.3 to 100.0, and 99.5 to 99.8 per cent., respectively (cf. Griebel and Weisz, *Z. Unters. Lebensm.*, 1935, **70**, 61; Deshusses, *Mitt. Lebensm. Hyg.*, 1939, **30**, 10).

J. G.

Colouring Matters of Flowers of *Tormentilla potentilla*. L. Schmid and A. Polaczek-Wittek. (*Mikrochem.*, 1939, **27**, 42–46.)—The flowers of *Tormentilla potentilla* contain a number of polyene colouring matters and two flavones and flavonols. In the tests described 5 g. of dried petals were used. The polyenes were identified by optical measurement of the positions of maximum absorption; β -carotene and lutein were recognised with certainty; two other spectra were probably due to zeaxanthine and flavoxanthine. In the saponifiable portion myristic acid was identified by micro-combustion, and an unsaturated acid was also present. The unsaponifiable matter contained paraffin hydrocarbons.

J. W. M.

New Colour Reaction of Phenarsazine Chloride. J. Delga. (*J. Pharm. Chim.*, 1940, **132**, 73–76.)—Phenarsazine chloride (Adamsite) gives in acetic acid solution and in presence of silver nitrate a yellow colour, and it is possible thus to detect 0.02 to 0.04 mg. To a small quantity of oxide or chloride of phenarsazine in a test-tube are added 5 ml. of the reagent (25 ml. of a 10 per cent. solution of silver nitrate and 25 ml. of glacial acetic acid), and after being kept for 10 minutes in a boiling water-bath the colour is noted. With 0.05 mg. of Adamsite this is a clear yellow becoming intense with 0.2 mg. Twenty other arsenic derivatives, including mono- and di-phenylchloroarsine, acridarsines and sodium cacodylate, gave no colour reactions. With chloride derivatives a curdled white precipitate of silver chloride was formed. Paranitrophenarsazine chloride alone dissolved to a yellow solution in acetic acid, but the colour was not intensified on warming, even in presence of silver nitrate. Diphenylamine, a constant impurity in industrial Adamsite, gave with the reagent a dirty green colour changing to black. To detect Adamsite in water, 0.25 g. of silver nitrate is added to 5 ml. of the suspected water and dissolved by shaking, after which 5 ml. of pure acetic acid are added and the mixture is left for 10 minutes in a boiling water-bath; if Adamsite is present the characteristic colours will be produced; one part of Adamsite in 125,000 of water may thus be detected.

D. G. H.

Inorganic

Separation of Cadmium from Zinc by Precipitation with Aluminium.

F. E. Townsend and G. N. Cade. (*Ind. Eng. Chem., Anal. Ed.*, 1940, **12**, 163-164.)—The acid sulphate solution (200 ml.), containing zinc and cadmium, is neutralised with sodium hydroxide and acidified with 1 ml. of conc. sulphuric acid. Granular aluminium (< 40 mesh, 0.3 to 1 g. according to the amount of cadmium present) is added, and the liquid is boiled for 5 minutes. The precipitate is filtered off on a wad of cotton-wool and washed with cold water, and the filter and precipitate are returned to the precipitation-beaker. One-tenth g. of aluminium is added to the filtrate, which is boiled for 5 minutes, and filtered through a filter-paper. The residue is washed with cold water, and the filter is added to the first one. The filters are macerated and treated with 5 ml. of sodium hydroxide solution (50 per cent.) to dissolve out metallic aluminium. After addition of water, the spongy cadmium which remains is filtered off and determined by any convenient method. Practically quantitative results were obtained in the separation of 0.1 g. of cadmium from quantities of zinc up to 2.0 g. Analysis of various metallurgical products with the aid of the method gave good results for cadmium in comparison with the hydrogen sulphide separation method. S. G. C.

Detection of Gold by Means of Morpholin. L. S. Malowan.

(*Z. anal. Chem.*, 1939, **118**, 100-102.)—Morpholin, C_4H_9NO , is a cyclic secondary amine, prepared from diethanolamine. It is a colourless liquid with an odour of ammonia and is miscible in all proportions with water (Wilson, *Ind. Eng. Chem.*, 1935, **27**, 867, 870; Malowan, *Mikrochemie*, 1939, **26**, 319; Abst., *ANALYST*, 1939, **64**, 765). Owing to its alkalinity it precipitates hydroxides of various metals; only cadmium forms an organo-compound. Morpholin can easily be oxidised and reduces metallic salts. Gold and silver are not precipitated in the cold, but only on boiling the solution. Silver gives a mirror, and gold first a yellow colour and then red-violet flakes. The reaction is obtained with as little as 1 ml. of a solution containing 1 part of gold in 50,000. For the test, 2 to 3 ml. of the solution are mixed with 0.3 ml. of pure morpholin, or such amount that the solution becomes distinctly alkaline. If copper or iron is present it is precipitated; the filtrate from the precipitate is boiled, and if gold is present the solution first shows a yellow colour, which changes gradually to bluish-violet, and finally bluish-violet flakes are precipitated. The reaction is very reliable and enables an approximate estimation of the amount of gold to be made. D. A.

Assay for Platinum Metals in Ore Concentrates. J. Seath and

F. E. Beamish. (*Ind. Eng. Chem., Anal. Ed.*, 1940, **12**, 169-171.)—Details of the dry assay of platinum metals in concentrates from nickel ores are discussed.

S. G. C.

Determination of Chromic Oxide in Leather. I. Comparison of Wet and Dry Oxidation Methods. J. C. Mertz. (*J. Amer. Leather Chem. Assoc.*, 1940, **35**, 36-43.)—Six commercial chrome-tanned leathers in strip form (including blue-, brown- and black-dyed, and undyed one- and two-bath calf, suede-calf and

kid leathers) were ground in a Wiley mill and analysed by the following methods:— (1) *Smith and Sullivan Method* (see ANALYST, 1935, 60, 779).—It was found preferable to use 2-g. samples and 0.1 N solutions. (2) *Modified American Leather Chemists' Association Method* (cf. Wilson and Merrill, "Analysis of Leather," 1931, p. 22).—The sample (3 g.) was ignited in a platinum dish, the ash was fused for at least one hour with 4 g. of a mixture containing equal weights of sodium carbonate, potassium carbonate and powdered borax glass, and the melt was extracted with an excess of dilute hydrochloric acid. A few drops of sulphuric acid were added to the acid solution, which was then boiled for 2 minutes. As no barium sulphate was found, the clear solution was diluted to 250 ml., and a mixture of 100 ml. of this solution with 5 ml. of conc. hydrochloric acid and 10 ml. of a 10 per cent. solution of potassium iodide was placed in a stoppered flask in the dark for 2 minutes. The iodine liberated was then titrated with 0.1 N sodium thiosulphate solution, 2 ml. of a 0.5 per cent. solution of starch being used as indicator. The iron and aluminium were removed from another 100 ml. of the solution in the usual way, and the chromium in the resulting combined filtrate and washings was determined as described above. (3) *Combination Method*.—The Smith and Sullivan method (*loc. cit.*) was followed up to, and including, the operations of diluting to 200 ml. and cooling. The solution was then diluted to 250 ml., and 100-ml. portions were taken for the iodimetric determination of chromium by Method (2), both with and without removal of iron and aluminium.

The results varied from 2.65 to 6.69 per cent. of Cr_2O_3 (dry basis), and those obtained by Methods (1) and (2) were in excellent agreement. Method (1), however, is preferred, as the wet oxidation eliminates the long fusion period (which must not be shortened), the appreciable losses of platinum from the crucible, and the interfering effect of contamination by platinum on the colour of the starch-iodine complex, which is changed to an amber-red shade. It is important, however, that in Method (1), 80 per cent. sulphuric acid should be used in the oxidising solution, since use of 95 to 98 per cent. acid without dilution will dehydrate the perchloric acid to an extent incompatible with safety. The iron present in the samples examined was sufficient to influence the results, and if it was not removed the results were high by 0.1 to 0.2 (in one test, 5) per cent. of Cr_2O_3 . Method (3) gave results that were consistently low, *viz.* by 3 to 10 per cent. of the chromium-content. This difference is attributed to the effect, on the iodimetric titration, of the anions introduced by the oxidising agents, and not to incomplete oxidation. J. G.

Determination of Molybdenum in Cast Iron and Steel. C. Sterling and W. P. Spuhr. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 33–34.)—Knowles's α -benzoinmonoxime method is employed (cf. ANALYST, 1932, 57, 799–801) with the modification that, instead of the precipitate being ignited to molybdic oxide, an operation found to be troublesome and necessitating corrections for impurities, the α -benzoinmonoxime-molybdenum precipitate is dissolved in ammonia and the molybdenum is precipitated as lead molybdate. The treatment is as follows: The α -benzoinmonoxime-molybdenum precipitate after filtration is washed back into the precipitation-beaker, the filter being reserved; 10 ml. of ammonia and 10 ml. of 30 per cent. hydrogen peroxide are added, and the liquid is diluted to

75 ml., and boiled until oxygen evolution ceases. The liquid is filtered through the original filter, which is then washed with hot dilute ammonia (1:50). The lead molybdate precipitation is effected by pouring the filtrate into 100 ml. of boiling lead acetate buffer mixture (4 g. of lead acetate crystals dissolved in a mixture of 550 ml. of ammonia (sp.gr. 0.90), 900 ml. of 50 per cent. acetic acid, 275 ml. of conc. hydrochloric acid and 275 ml. of water). The liquid is boiled, the precipitate is allowed to settle for 30 minutes (a longer period is advised for small precipitates), filtered off on a close-textured paper, and washed with hot 2 per cent. ammonium acetate solution containing a little acetic acid. The filter is ashed, and the precipitate is finally ignited at a dull red heat. The method is applicable to tungsten-free molybdenum-bearing steels and irons. Tungsten, tantalum and niobium interfere with the α -benzoinmonoxime precipitation method. With tungsten present, preliminary separation of the molybdenum as sulphide is advised (*cf.* Arrington and Rice, *Bur. Mines Rept. Investigations*, 3441, pp. 39-59, 1939).
S. G. C.

Determination of Cerium by Means of 8-Hydroxyquinoline. R. Berg and E. Becker. (*Z. anal. Chem.*, 1940, 119, 1-4.)—The neutral solution, containing 0.01 to 0.05 g. of cerium, is warmed with 1 g. of hydroxylamine hydrochloride until colourless, treated with excess of sodium tartrate and 20 ml. of 2 *N* ammonia, diluted to 100 ml., and treated at 60° C. with a 2 per cent. alcoholic solution of the reagent until the supernatant liquid is orange-yellow. It is then heated to boiling and left for 30 minutes over a small luminous flame. The precipitate is collected in a porous glass crucible (G 4) or on an ashless filter, and washed with warm feebly ammoniacal water until the washings are colourless, and weighed after having been either dried at 110° C. or ignited to CeO_2 under a layer of sublimed oxalic acid (Ce factor for $\text{Ce}(\text{C}_8\text{H}_6\text{ON})_3$: 0.2447). The precipitate may also be treated bromometrically (*ANALYST*, 1934, 59, 325). For the separation of thorium from cerium, Hecht and Ehrmann's hydroxyquinoline method for the determination of thorium is applicable (*ibid.*, 1935, 60, 272), 1 g. of hydroxylamine hydrochloride being added (*supra*). The filtrate from the thorium precipitate is treated with sodium tartrate, etc., as described above.
W. R. S.

Volumetric Determination of Tungsten. M. L. Holt and A. G. Gray. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 144-146.)—The method is based on the reduction of sexavalent to tervalent tungsten by liquid lead amalgam in a specially designed reductor. *Apparatus.*—The reductor consists of a 400-ml. tap funnel with a stopper carrying a leading tube for admission of carbon dioxide and a smaller tap-funnel for admitting liquid. The reductor-funnel is provided with an exterior winding of nichrome wire covered with asbestos cement for electrical heating of the contents. The stem of the reductor-funnel has a two-way tap with two leading tubes, one to act as a run-off and the other communicating with the titration bottle. The titration bottle is a three-necked Woulff bottle; the three necks are stoppered and carry respectively (1) the tube from the reductor, (2) burette, (3) tap-funnel for admission of liquid; a side-tube entering near the bottom of the Woulff bottle carries a tube for admission of carbon dioxide for the purpose of agitating the liquid during titration. *Lead amalgam.*—A mixture of 40 g. of

granulated lead and 600 g. of mercury is heated at 100° C. for 1 hour with constant stirring. The amalgam is cooled and filtered through a fine cloth filter to remove the solid portion, which is rejected. The liquid amalgam is washed with water and dried with filter-paper. *Method.*—The test solution, containing the tungsten as sodium tungstate, is evaporated almost to dryness in a separate vessel; 75 ml. of conc. hydrochloric acid are added; the liquid is heated to produce a clear solution, and introduced into the reductor, from which air has previously been swept by carbon dioxide. Lead amalgam is run in, and the reductor is gently shaken at 60° until the solution has passed through various colour changes and reached a deep yellow colour; reduction is continued for a further 5 minutes (about 15 minutes in all) to ensure completeness. The amalgam is then run off through the two-way tap. A slight excess of ferric iron solution (10 per cent. ferric ammonium sulphate solution in 25 per cent. phosphoric acid) is added to the reduced tungsten solution, and the mixture is run off into the titration bottle together with 10 to 20 ml. of water used for rinsing out the reductor; 75 ml. of water, 40 ml. of syrupy phosphoric acid and a few drops of diphenylamine sulphonc acid indicator solution are added to the contents of the titration bottle, and the solution is titrated with *N*/10 dichromate solution. The calculation of the result is based on the change of valence of tungsten from 3 to 6. Results close to the theoretical were obtained in tests with 0.003 to 0.16 g. of tungsten. The method was applied to the analysis of electroplated tungsten-nickel alloys and to ferro-tungsten, the tungsten being first obtained in solution as sodium tungstate by normal methods.

S. G. C.

Determination of Uranium and Copper with the aid of the Silver Reductor. N. Birnbaum and S. M. Edmonds. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 155–157.)—*Uranium.*—The solution (50 ml.), containing 0.1 to 0.4 g. of uranyl salt in 4 *M* hydrochloric acid, is heated to 60° C., and passed at the rate of 20 ml. per minute through a silver reductor which has been pre-heated by the previous passage of hot 4 *M* hydrochloric acid. (The silver reductor is described by Walden, Hammett and Edmonds, *J. Amer. Chem. Soc.*, 1934, 56, 350; cf. Fryling and Tooley, *id.*, 1936, 58, 826, Abstr., ANALYST, 1936, 61, 722). The reductor column is washed through with 150 ml. of hot 4 *M* hydrochloric acid. Uranium is reduced to the quadrivalent state. The reduced solution is cooled, 3 ml. of 85 per cent. phosphoric acid and 1 drop of *o*-phenanthroline ferrous-complex indicator solution are added, and the solution is titrated with 0.1 *M* ceric sulphate solution. Acetic acid does not interfere. Nitric acid, iron, molybdenum, vanadium and copper, which are also capable of reduction, must be absent. *Copper.*—The solution (50 ml.) containing 0.1 to 0.4 g. of copper in 2 *M* hydrochloric acid is passed through the reductor at 25 ml. per minute (temperature of solution not stated). The reduced solution, containing cuprous chloride, is collected under 20 ml. of 0.5 *M* ferric alum solution. The reductor column is washed through with 150 ml. of 2 *M* hydrochloric acid, and the solution is titrated with 0.1 *M* ceric sulphate solution, 1 drop of *o*-phenanthroline complex being added as indicator. Nitric acid (up to about 5 per cent. strength) is without effect on the accuracy.

S. G. C.

Detection of Magnesium by means of *p*-Nitrobenzeneazoresorcinol.**J. P. Mehlig and K. R. Johnson.** (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 30-31.)—

The effect of forty-four of the common ions upon the *p*-nitrobenzeneazoresorcinol test for magnesium has been studied. To the solution, containing 10 mg. of magnesium and up to 1000 mg. of the ion under test, 1 drop of reagent solution (0.5 g. of *p*-nitrobenzeneazoresorcinol dissolved in 100 ml. of 1 per cent. sodium hydroxide solution) is added, and the solution is rendered alkaline with sodium hydroxide. Magnesium yields a sky-blue precipitate; sensitiveness 0.002 mg. in 10 ml. Interference is caused by the ions forming coloured precipitates masking the magnesium blue. Black precipitates are given by mercurous and mercuric ions; brown by silver, manganous, ferric and arsenate ions; green by chromic, nickelous, and ferrous ions; yellow by chromate and blue by cupric ions. Only arsenate, chromate and permanganate, out of the 20 anions tested, interfered. The white precipitates formed by aluminium, antimonious, bismuth, cadmium, stannic, stannous and zinc ions do not interfere, as no masking of the blue is produced. Ammonium salts cause no trouble unless present in considerable excess. The method is considered advantageous for use in the "group" detection of magnesium; interfering ions are removed in the course of the prior separations. Aluminium, barium, calcium and strontium, while tending to interfere with the traditional magnesium ammonium phosphate test, are without effect on the present method.

S. G. C.

Determination of Bromides in Presence of Chlorides. A. Denoël.

(*J. Pharm. Belg.*, 1940, 22, 179-184.)—The principle of the method (which has the advantages of accuracy and rapidity) is the liberation of bromine from the bromides by the action of potassium bromate under specified conditions of acidity, the chlorides being unattacked. The bromine is then extracted with carbon tetrachloride, and allowed to liberate iodine from potassium iodide; the former may be titrated. To a mixture of 100 ml. of the sample (which should not contain more than 0.1 g. of bromide ion), with 8 ml. of 10 per cent. sulphuric acid and 10 ml. of 5 per cent. potassium bromate solution, are added 50 ml. of carbon tetrachloride. The mixture is shaken well, and after 10 minutes the carbon tetrachloride layer is transferred to a stoppered 500-ml. flask containing 50 ml. of a 4 per cent. potassium iodide solution. Four further extractions, each with 40 to 50 ml. of carbon tetrachloride, are made; the last should remain colourless. The combined extracts are then shaken in the flask, and the liberated iodine is titrated with 0.1 *N* sodium thiosulphate solution, the end-point being the simultaneous disappearance of the violet colour of the carbon tetrachloride and of the yellow colour of the aqueous layer. If the chloride-content is required, a preliminary determination of the total halogens must be made by the Volhard-Charpentier method and the bromide-content deducted; it is thus possible to determine 1 per cent. of potassium chloride in potassium bromide, or *vice versa*. The method is specific for bromides in absence of iodides, and is unaffected by the presence of up to 0.3 g. of sodium chloride. It is shown that no acid or bromate is carried over with the carbon tetrachloride, but a blank test on the reagents is advised. Tests with solutions of pure potassium bromide containing 3 to 100 mg. of bromide ion gave results that

were 0.7 per cent. low, but agreed well with those obtained by Charpentier's method and by gravimetric analysis. In presence of up to 0.3 g. of sodium chloride the recorded error varied from -0.69 to $+1.03$ per cent., but higher chloride-contents produced greater errors. When the quantity of bromide is very small the best results are obtained by the use of 100 ml. of water, 5 ml. of 10 per cent. sulphuric acid and 5 ml. of 5 per cent. potassium bromate solution. With organic compounds the possibility exists that nitrites formed by ignition in presence of nitrates will reduce some of the bromate to bromide and so give high results. It is not advisable to destroy the nitrites by means of potassium permanganate, and it is preferable to destroy the organic matter by ignition in presence of sodium carbonate.

J. G.

Argentometric Determination of Cyanide with Diphenylcarbazone as Indicator. R. Ripan-Tilici. (*Z. anal. Chem.*, 1940, 118, 305-307.)—The cyanide solution (0.01 to 0.05 *M*) is treated with 4 or 5 drops of an alcoholic 0.3 per cent. solution of the indicator, which colours it reddish-brown, and titrated during agitation with 0.01 to 0.05 *M* silver nitrate solution. When all of the cyanide has been converted into the complex $[\text{Ag}(\text{CN})_2]^-$, a further drop of silver solution turns the liquid violet. If now the titration is continued, the intensity of the violet colour increases, until one drop of silver solution colours the precipitate blue and causes it to flocculate. The total volume of silver solution thus consumed is twice that required for the production of the violet end-point, where the diphenylcarbazone acts as a colour indicator for the ratio $2\text{CN}:\text{Ag}$; at the blue end-point, it acts as an adsorption indicator for the ratio $\text{CN}:\text{Ag}$. Both end-points are accurate and sharp (*cf.* ANALYST, 1935, 60, 428).

W. R. S.

Determination of Free Cyanide in Brass Electroplating Baths. W. M. McNabb and S. Heiman. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 161-163.)—The authors' work indicates that a brass-plating solution contains $\text{NaCu}(\text{CN})_2$, $\text{Na}_2\text{Cu}(\text{CN})_3$, $\text{Na}_2\text{Zn}(\text{CN})_4$, Na_2ZnO_2 , free NaCN , with small amounts of amines and alkali. The equilibrium among these constituents, and consequently the value of the free cyanide, is shifted by changes in the concentration of the constituents, the temperature and the *pH* value. Nevertheless it is possible to determine the quantity of "free cyanide" required to yield results which, though empirical, are useful for the purpose of controlling the operation of the bath. The most satisfactory method would appear to be that of Pan (*Metal Industry*, New York, 1932, 30, 402-404), which is as follows:—To 10 ml. of the brass-plating solution are added 4.3 g. of potassium iodide and sufficient water to produce a volume of 70 ml. at the end of the titration. The solution is titrated with *N*/10 silver nitrate solution to the appearance of a bluish opalescence. The free sodium cyanide, in g. per litre, is given by multiplying the number of ml. of *N*/10 silver nitrate by 0.980.

S. G. C.

Determination of Silica in Cryolite and Materials containing Fluorine. H. Spielhacsek. (*Z. anal. Chem.*, 1940, 119, 4-16.)—A new method is described in which the ore is fused with bisulphate, volatilisation of silicon being prevented

by addition of borax, with the result that the fluorine escapes as boron trifluoride. One g. of ore is mixed with 3 g. of fused (or 6 g. of crystallised) borax and 14 g. of powdered potassium bisulphate in a platinum crucible. The covered crucible is cautiously heated round the sides until the reaction is subsiding, then more strongly until quiet fusion is attained. The cooled crucible is heated with 100 ml. of water and 3 ml. of hydrochloric acid in a porcelain basin, carefully cleaned, and the liquid is evaporated to the consistency of a syrup. After cooling, the crystalline mass is crushed with a flattened glass rod, the basin is heated in an oven at $110^{\circ}\text{C}.$, and after renewed cooling the dry mass is thoroughly pulverised. It is then returned to the oven and heated until hydrochloric acid can no longer be detected by its odour, cooled, moistened with hydrochloric acid, and taken up with hot water after a few minutes' standing. After digestion on a water-bath the solution is filtered through close-textured paper [a pad of filter-pulp in the filter would be advisable—*Abstractor*], the precipitate is washed free from sulphate, heated in a tared platinum crucible, and ignited to SiO_2 . This is tested for purity with hydrofluoric acid as usual. A blank test is advisable. The method is simple, more rapid and reliable than other procedures, and specially suitable for serial determinations. The results obtained on synthetic mixtures are very satisfactory.

W. R. S.

Microchemical

New Method for Washing and Isolation of Barium Sulphate in the Micro-Determination of Sulphur. R. Grangaud. (*Mikrochem.*, 1939, 27, 52–56.)—After decomposition, by the micro-Carius or Pregl method, of the organic substance containing sulphur, the precipitation is carried out in a Jena glass tube in the usual way, the liquid is evaporated to dryness, and the residue is stirred up with 1 per cent. hydrochloric acid and transferred to a weighed quartz test-tube by rinsing alternately with alcohol and 1 per cent. hydrochloric acid. After centrifuging, the supernatant liquid is drawn off by suction, a tube bent round to turn upwards being used to avoid disturbing the precipitate. The precipitate is washed 3 times with 1 per cent. hydrochloric acid and centrifuged after each washing. Finally the test-tube is dried, heated to redness, cooled and weighed.

J. W. M.

Colorimetric Micro-method for the Estimation of Sodium with Manganous Uranyl Acetate. E. Leva. (*J. Biol. Chem.*, 1940, 132, 487–499.)—The whole blood, serum or urine is first deproteinised by introducing 0.10 or 0.20 ml. into a 5-ml. flask containing 0.5 ml. of 20 per cent. trichloroacetic acid. The solution is diluted to the mark with water and thoroughly shaken, the foam being broken by touching the surface with a wire dipped in capryl alcohol. With protein-free materials, the trichloroacetic acid treatment may be omitted. One-ml. aliquot portions of the filtered solution are transferred to 15-ml. centrifuge tubes, 9 ml. of 25 per cent. alcoholic manganous uranyl acetate reagent are added to each, and the solutions are mixed with a clean glass rod. (This reagent is prepared as follows: To 160.0 g. of $\text{UO}_2(\text{CH}_3\text{COO})_2 \cdot 2\text{H}_2\text{O}$, 490.0 g.

of $\text{Mn}(\text{CH}_3\text{COO})_2 \cdot 4\text{H}_2\text{O}$ and 138.0 ml. of 30 per cent. acetic acid, 1292.0 ml. of water are added. The manganous acetate should be in the form of pink lustrous crystals, free from greyish powder. The salts dissolve if the flask is shaken occasionally, and a precipitate of the sodium salt separates out and settles after a while, leaving a clear solution. To prepare the reagent from this stock solution, 9 ml. are mixed with 3 ml. of 95 per cent. alcohol, and the mixture is allowed to stand in the dark for at least 4 hours and then filtered. The reagent keeps for about 3 weeks if stored in the dark.) A precipitate is formed in about a minute and the rod is then rinsed with a further 1 ml. of the reagent and removed. The centrifuge tubes are covered with rubber caps and allowed to stand in the dark for at least 4 hours, after which they are centrifuged and the supernatant liquid is discarded. The precipitate and the walls of the tubes are washed free from manganese by adding 4 ml. of washing solution from a pipette, and suspending the precipitate in this solution by stirring with a fine glass rod, which is then rinsed with 1 ml. of washing solution and removed. (The washing solution is made as follows: To 160.0 g. of $\text{UO}_2(\text{CH}_3\text{COO})_2 \cdot 2\text{H}_2\text{O}$, 440.0 g. of $\text{Zn}(\text{CH}_3\text{COO})_2 \cdot 4\text{H}_2\text{O}$, and 138.0 ml. of 30 per cent. acetic acid, 1342.0 ml. of water are added, the salts being dissolved in the same way as in the preparation of the manganous uranyl reagent. If insufficient sodium is present to saturate the solution, 0.1 N sodium chloride solution should be added until a precipitate persists. Twelve ml. of this solution are mixed with 4 ml. of 95 per cent. alcohol, and the solution is saturated with manganous triple salt by adding 20 mg. of the solid salt to each 100 ml. of solution. The solution is allowed to stand for 1 hour or longer and is then filtered; it keeps for 3 weeks in the dark. The triple salt is prepared by adding 125 ml. of the stock solution of manganous uranyl acetate to 2 ml. of a 5 per cent. sodium chloride solution. The mixture is stirred and allowed to stand for half-an-hour, when the supernatant liquid is decanted. The precipitate is transferred to a 50 ml. centrifuge tube and centrifuged, and the supernatant liquid drawn off. The precipitate is washed by suspending and centrifuging three times with 95 per cent. alcohol and twice with ether. The triple salt is dried and stored in a brown tube.) The tubes are again centrifuged and the washing is repeated twice. For the third washing it is unnecessary to re-suspend the precipitate, 4 to 5 ml. of the washing solution being simply added to the tubes. These are then placed in water, the temperature of which is raised slowly to the boiling-point, and most of the remaining supernatant liquid is evaporated off. Ten ml. of potassium periodate solution (2.5 g. dissolved in about 400 ml. of water, and the solution treated with 100 ml. of 85 per cent. phosphoric acid, shaken and made up to 1 litre with water) are introduced into each of the tubes, the contents of which are stirred with a glass rod, and they are then immersed in boiling water for 10 to 15 minutes. After cooling, the solutions are transferred quantitatively to graduated flasks, 25 or 50 ml. for 0.10- or 0.20-ml. samples, respectively. The solutions are diluted to the mark and mixed. The sodium concentrations are estimated either by comparing the colours with standard potassium permanganate solution (0.2200 g. of dry potassium permanganate is dissolved in water, 200 ml. of the potassium periodate reagent are added, and the solution is diluted to 1 litre with water; 4 ml. of this solution are then diluted to 50 ml. with water), or by measuring the extinction value in an

Evelyn photoelectric colorimeter, filter 520 being used. Using the former method, the concentration of sodium is calculated from the equation

$$\frac{20 \times \text{mM. KMnO}_4 \text{ in standard} \times 1000}{\text{Reading} \times \text{ml. of sample precipitated}} = \text{mM* sodium per litre}$$

the standard being set at 20 mm. When the photoelectric colorimeter is employed, the calculation is made by the use of the equation

$$\frac{\text{Density (from the galvanometer reading)} \times 1000}{39.27 \times (100/\text{volume of oxidised solution}) \times \text{ml. of sample analysed}} = \text{mM* sodium per litre}$$

39.27 being the value of the density of 1 mM potassium permanganate solution per 100 ml. for the particular instrument used in the investigation; it should be re-determined for each laboratory. The presence of phosphate, even in equimolecular proportions, does not affect the accuracy of the sodium estimations, the values for sodium varying by not more than ± 1.5 per cent. from the theoretical. Potassium causes high results if the molar ratio K : Na in the sample exceeds 1.5. Excess of potassium may be removed by precipitation as the perchlorate. The results obtained by the method (with either method of measurement) were in close agreement with the results obtained by the gravimetric method of Butler and Tuthill (*J. Biol. Chem.*, 1931, 93, 171; cf. *ANALYST*, 1931, 56, 764).

F. A. R.

* Milli-moles.

Reviews

SULPHATED OILS AND ALLIED PRODUCTS.—THEIR CHEMISTRY AND ANALYSIS.

By DONALD BURTON, M.B.E., D.Sc., F.I.C., and GEORGE F. ROBERTSHAW, A.M.S.T., A.I.C. With a Foreword by Prof. T. P. HILDITCH, D.Sc., F.I.C. Pp. iv + 163. London: A. Harvey. 1939. Price 12s. 6d. net.

Many different oils react with sulphuric acid, and each yields a wide variety of products whose nature depends on the conditions employed. The reaction, first studied over a hundred years ago, found early technical application in the use, by Runge in 1834, of sulphated olive oil as a mordant in dyeing calico red. Since then it has been extensively developed and many sulphated products are now used in leather manufacture, in the textile industry, as detergents, wetting agents and machine oils, and even in food manufacture.

As Professor Hilditch points out, our knowledge of the chemistry of the process of sulphation is still very incomplete, partly because the reactions are of great complexity and partly because the analytical chemistry, perforce restricted to determinations of general "characteristics," is not always easy to interpret.

Varying technique in analysis formerly led to much confusion, but the position has been considerably clarified by international collaboration, in which the

authors of this monograph have taken a prominent part. A scheme of analysis was put forward in 1931 and 1933, and the present work is the result of its cordial reception and requests for a revised edition.

The book includes a brief historical survey of the subject and descriptions of the raw materials used and of the chemistry and methods of sulphation. Most of it is devoted to three chapters dealing with the analysis of sulphated oils, sulphated fatty alcohols and petroleum sulphonc acids. In these three chapters there are excellent descriptions of methods drawn from widespread sources, and the practical notes appended to each description are of the greatest value as a guide to the interpretation of results and as a warning of the limitations and difficulties yet to be overcome.

The survey of the present position of the analytical chemistry of the subject is complete and critical and will provide an immediate stimulus to further progress. The authors have rendered a great service in collecting and presenting in so clear a manner all the analytical information available about this complex subject.

K. A. WILLIAMS

THE CHEMICAL CONSTITUTION OF NATURAL FATS. By T. P. HILDITCH, D.Sc., F.I.C. Pp. x + 438. London: Chapman & Hall, Ltd. 1940. Price 35s. net.

In the year 1823 Chevreul announced his discovery that natural fats consist of the glycerol esters of certain acids, among which palmitic, stearic and oleic are prominent; this book is the logical outcome of this discovery, but it would appear desirable to consider the reasons which have caused so long a time to elapse between the discovery of the raw material and the issue of the "finished product."

For years the discovery of Chevreul failed to attract the attention of a research school, and the little progress that was made was due to the work of isolated observers, who considered the matter as a hobby rather than as the serious work of a lifetime. The apparent difficulty of the subject, together with the existence of what seemed to be more attractive fields for investigation, did not add to the favour with which the subject was regarded. It is now, of course, well known that the presence of simple triglycerides in a fat is the exception rather than the rule, and that they are only present when the formation of mixed triglycerides is impossible owing to the presence of a predominating proportion of one acid; yet nearly eighty years after Chevreul's discovery, the presence of a mixed triglyceride in a fat was described as "unusual."

During the last fifteen to twenty years, however, our knowledge of the chemistry of natural fats has been revolutionised. A number of institutions have taken up the work in a thorough and convincing manner. In the forefront we have the Liverpool School, ably led by Professor Hilditch, which has been responsible for so many of the new devices, and improvements of the older devices, and which has played so large a part in the progress made.

In eleven chapters Professor Hilditch unfolds the mechanism of attack and the vast number of results obtained. After an introductory chapter of 21 pages, three chapters (occupying 159 pages) deal with the constituent acids found in many

hundreds of natural fats obtained from the simplest to the highest organisms. Professor Hilditch is careful to point out that our knowledge is by no means complete, but even a superficial comparison of these pages with books published less than twenty years ago will show the tremendous advances made during this period.

The next three chapters (81 pages) deal with the constituent glycerides of natural fats, this being the first time that a comprehensive account of this division of the subject has been attempted. Chapter VIII deals with some aspects of the biochemistry of fats, Chapter IX with the constitution of individual naturally occurring fatty acids, and Chapter X with synthetic glycerides. The final Chapter, XI, gives an account of the experimental methods now used in investigating the constitution of fats. Most of these methods have either been devised or elaborated at Liverpool, so that the working details here given can be regarded as authentic.

It is not often that a worker who has made wide and important advances in a particular subject can be persuaded to write a book giving the results obtained by himself and by others, and the task has perforce to be undertaken by those less qualified to do so. Our debt to Professor Hilditch is therefore the greater because, not only has he successfully completed a labour of great magnitude, but readers can be satisfied that the whole array of facts has been presented by one who has personal knowledge of the details of the subject on which he writes.

This work will be literally indispensable to all those who require a knowledge of the composition of natural fats. Paper, printing and binding are of satisfactory quality, and help to produce a book of which the author may well be proud.

G. D. ELSDON

LAVOISIER. By J. A. COCHRANE. Pp. xii + 264, 9 illustrations. London: Constable & Co. 1939. Price 3s. 6d. net.

This is a cheap edition of a work first published in 1931. The author mentions the fact that Lavoisier, at his death, was, and had been for fifteen years previously, one of the most eminent men in France; yet the general historian does not usually think it worth while even to mention him. Among chemists he has been honoured from the day of his death as a chief founder of modern chemistry; yet even they are less acquainted with his career as a whole than might have been expected. We are thoroughly well informed about his discovery of oxygen and his routing of the phlogisticians, as well as of the work he did in connection with the devising of a new nomenclature of chemistry, but we are not nearly so familiar with other aspects of his life and work in relation to the times and conditions in which his lot was cast. We have imbibed something of his scientific spirit from his own *Traité élémentaire de Chimie*, wherein he stated: "Il n'est jamais permis, en physique et en chimie, de *supposer* ce qu'on peut *déterminer* par des expériences directes"; we know, too, with what contempt he was treated by Coffinhal at his trial, when he was told that "The Republic has no need of savants."

Mr. Cochrane not only discusses fully the main facts of Lavoisier's chemical work and writings familiar to all interested in the history of the science, but also

deals at length with his very varied work in connection with the numerous investigations he undertook on behalf of the Academy of Sciences. We have here provided for us a picture of this versatile man of science seen in the perspective of events and personalities which exhibit him as a truly great man suffering obloquy and death because of an eminence begotten of his disinterested labours on behalf of his fellow citizens. The book contains chapters on Domestic Affairs, Politics, Personal Attacks, National Education, Arrest, In Prison, and The Guillotine. The illustrations are especially satisfactory. It is altogether a very cheap and desirable volume.

W. KIRKBY

DISCOVERY OF THE ELEMENTS. By MARY ELVIRA WEEKS. Fourth Edition; enlarged and revised. Pp. viii + 470, with many illustrations. Published by *Journal of Chemical Education*, Easton, Pennsylvania, U.S.A. 1939. Price \$4.00 post free.

The fact that a fourth edition of this book has been called for so soon is, perhaps, its best recommendation to the reader, as few books dealing with the history of science can claim such distinction. Its popularity is not difficult to understand, since the author makes an interesting story out of what others might record in a dry-as-dust manner, yet achieves this without any departure from scholarship. Not only are the researches leading to the first recognition and isolation of the various elements described with a wealth of detail, quotation from original papers and ample bibliographical references, but short biographical accounts are also given of the persons most closely connected with these discoveries. In addition, the many illustrations, portraits and reproductions of MSS., collected from many sources by Professor Dains, make the book attractive to the chemist interested in the human side of his subject.

The least satisfactory portions of the book are the first two chapters, on the elements known to the ancient world and elements known to the alchemists, which, with the exception of a good account of the discovery of phosphorus, are little more than fragmentary. In this section, too, the author does not show the judgment exercised in the latter portion of the book; for example, quotations from the Authorised Version of the Bible cannot be used as evidence of the scientific knowledge of the ancients. Perhaps this is best borne out by the passage cited in connection with allusions to iron in the Bible: "Oh that my words were now written! Oh, that they were printed in a book. That they were graven with an iron pen. . . ." (*Job*, xix. 23 and 24). If one accepts this as evidence of the use of iron for making a writing or engraving implement, one must assume that the printing press was in use in that era.

The additions to the book are becoming essays in historical chemistry rather than accounts of the discovery of the elements, so, perhaps, in future editions the sections devoted to classical and mediaeval knowledge of the elements will be elaborated. Although for the taste of some scientific readers the author perhaps shows rather too great a tendency to sentimentalise, the book is a mine of information on its subject and can be thoroughly recommended. O. L. BRADY

RECORDS AND RESEARCH IN ENGINEERING AND INDUSTRIAL SCIENCE. By J. EDWIN HOLMSTROM. Pp. 302. London: Chapman & Hall. 1940. Price 15s.

The scope of this work is described by the sub-title—"A Guide to the Production, Extraction, Integrating, Storekeeping, Circulation and Translation of Technical Knowledge." In these days of systematic record of the results of research and experiment, the whole of knowledge should be available to everyone, but it is not always easily accessible. In addition to about 14,000 books on technical and scientific subjects published annually, upwards of 750,000 articles and papers appear in about 14,000 technical and scientific journals. The systematic indexing and recording of all this material is a complicated and difficult business, but one of supreme importance if the accumulated knowledge is to be made readily available.

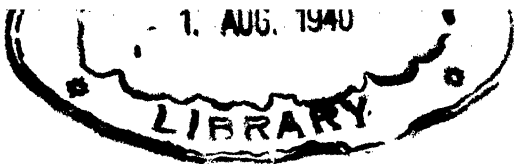
Dr. Holmstrom deals with the "Nature and Methods of Technical Science" and "Phases in the Application of Science to Practice," and then proceeds to discuss the work of Experimental and Collative Organisations. He describes in detail various methods of indexing, including the Universal Decimal Classification and the Kaiser system, as well as the facilities afforded by Bibliographical Bureau Services—not so well known as they deserve to be. No one particular system will suit every need, but the reader will find much useful and stimulating information in the details of the author's own system evolved to suit his particular problems. On a large scale the indexing and filing of technical information requires trained staff, especially in dealing with the problem of overlapping fields. This particularly thorny problem is fully discussed.

A chapter headed "The Expression and Transmission of Ideals" contains practical details of the mechanical processes involved in writing, dictating, type-writing and duplicating, as well as photographic reproduction and micro-photography, and on dealings with printers and publishers. The application of micro-photographic methods to collative research in reducing the bulk (by 99 per cent.) of literary material, both for more convenient circulation and storage, probably constitutes the most revolutionary advance in documentation since the invention of printing. America is ahead of Britain in the technique, but it is a satisfaction to record that a micro-film service is about to be initiated in this country at the small cost of about $\frac{1}{4}$ d. per quarto document. A reduction of 60 diameters would enable a 500-page book to be reproduced on a single 5×3 inch film for filing like a card index.

Another chapter contains much practical information on the Translation of Foreign Languages and Technical Dictionaries, especially as to the pitfalls that lie in the path of the unwary translator, usually more expert in his subject matter than in the language translated. Finally, the author touches on the possibilities offered by careers in occupations dependent on technical knowledge, with notes on specialisation and the employment of leisure time.

Essentially a practical work for practical men, Dr. Holmstrom's book fills a gap. It will be found particularly useful to the research worker and technician, who wishes to keep abreast of his subject, to know where to look for information, to obtain quick reference to original records and thus to avail himself to the full of the technical and scientific knowledge available.

R. W. SLOLEY



THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

AN Ordinary Meeting of the Society was held at 5.30 p.m., on Wednesday, May 1st, 1940, at the Chemical Society's Rooms, Burlington House, the President, Dr. E. B. Hughes, in the chair.

Sir William Willcox, K.C.I.E., C.B., C.M.G., F.I.C., was elected an Honorary Member of the Society, and J. A. Freeman, B.Pharm., B.Sc., F.I.C., Ph.D., M.P.S., and J. T. Stock, B.Sc., were elected Ordinary Members.

Applications for membership were read in favour of J. N. Davidson, B.Sc., M.D., J. R. Fraser, B.Sc., F.I.C., G. N. Gee, D. E. Jones, M.Sc., F.I.C., F. L. Kinsella, R. K. Matthews, F.I.C., W. J. Puregger, R. E. Stuckey, B.Sc., A.I.C., Ph.C., and J. A. C. Watson, A.I.C.

The following papers were presented and discussed:—"The Spectrophotometric Assay of Vitamin A, with special reference to Margarine," by J. R. Edisbury, B.Sc., Ph.D.; "The Estimation of Vitamin D in Margarine," by N. T. Gridgeman, B.Sc., H. Lees, B.Sc., and H. Wilkinson, B.Sc., Ph.D.; "The Composition of some Jam Fruits and the Determination of the Fruit Content of Jams," by T. Macara, F.I.C., and C. L. Hinton, F.I.C.

NORTH OF ENGLAND SECTION

A MEETING of the Section was held in Manchester on April 13th, 1940. The Chairman (Mr. J. R. Stubbs) presided over an attendance of thirty, which included the President (Dr. E. B. Hughes).

The following papers were read and discussed:—"The Iodimetric Determination of Alkali—An Investigation of Smith's Proposed Method," by J. Haslam, M.Sc., F.I.C., and R. F. Roberts; "Note on an Unusual Case of Cattle Poisoning," by J. G. Sherratt, B.Sc., F.I.C.

SCOTTISH SECTION

AN Ordinary Meeting of the Section was held in the North British Station Hotel Edinburgh, on Friday, April 19th, at 6.30 p.m.

The following papers were read and discussed:—"Notes on the use of Selective Oxidation in the Analysis of Fats," by W. A. Alexander, B.Sc., A.I.C.; "The Determination of Ketone Bodies in Body Fluids." "The Determination of Magnesium in Body Fluids," by H. Dryerre, Ph.D., M.R.C.S., L.R.C.P.; "The Pollution of Water Supplies by Trout and Gulls," by A. Dargie, B.Sc., A.I.C.

Obituary

SIR GILBERT THOMAS MORGAN, F.R.S.

THE death of Sir Gilbert Morgan on February 1st has removed one of the most distinguished of British chemists, and our Society deplores the loss of one of its Honorary Members. Many of us deeply regret the passing of an old and valued friend of many years standing, a man to whom one could go for sound advice and with whom one could talk freely on chemical and more mundane matters.

Born in 1870, Morgan was educated at the Central Foundation School and then at the Finsbury Technical College, where so many of our chemical friends received their scientific training. At that time the head of the Chemical Department was Professor Meldola, and the routine laboratory work was in the charge of J. Castell Evans and F. W. Streatfeild. Meldola had left the dyestuff industry for an academic career, and the research work at Finsbury was confined almost entirely to aromatic compounds containing nitrogen. Morgan assisted Meldola and Streatfeild in their work on the structure of the diazoamino-compounds, and after leaving College spent some time in the works of Read, Holliday & Co., at Huddersfield.

Returning to London, he was a demonstrator and then Assistant Professor at the Royal College of Science, where, at that time, Tilden was Professor of Chemistry. Always a great worker, Morgan found time to act as an abstractor, and afterwards edited the *Transactions of the Chemical Society* from 1903 to 1906.

Research work occupied all his spare time, and with the Finsbury tradition and the subsequent works experience it is no wonder that his earlier independent work was chiefly confined to aromatic compounds containing nitrogen. In conjunction with several co-workers, especially Miss F. M. G. Micklethwait, he published a series of papers, several of which dealt with diazoamino compounds and non-aromatic diazonium salts; the latter continued to interest him for some time after he left South Kensington. Morgan's interest was also aroused by the "residual affinity" of coumarin in forming salts with inorganic acids, and further by organic compounds in which arsenic and antimony were directly linked with carbon; both of these interests led to a large output of research in later years.

From 1912 to 1915 Morgan was Professor at the Royal College of Science, Dublin, and after a short period at British Dyestuffs, Ltd., succeeded Meldola as Professor of Chemistry at Finsbury Technical College, where he remained from 1916 to 1919. Publication was necessarily restricted during the war years, as he was engaged in matters of immediate importance for the country. His advice, meanwhile, was at the disposal of others, and the present writer had particular reason for gratitude in the matter of an autoclave of laboratory dimensions which was safe to a reasonably high pressure. Such articles were not often found in scientific laboratories in 1914!

In 1919 he became Mason Professor of Chemistry at Birmingham University, and a time of great activity followed. A crowd of enthusiasts was collected and worked from morning to night; even lunch was taken in the laboratory, and Morgan and his co-workers ate, drank coffee and "talked shop." The present writer has a happy memory of a casual visit to Birmingham and a lunch with these energetic chemists.

Arsenic and antimony linked to carbon had given another direction to Morgan's work and, not unnaturally, the corresponding sixth-group elements, selenium and tellurium, were soon under examination. Moreover, the "residual affinity" question had interested him in co-ordination, with the result that the different lines of research became merged and a large amount of work on metallic derivatives of various organic compounds was the result. The writer believes he is not mistaken in attributing the term "chelation" to Morgan.

In 1925 Morgan became Director of Chemical Research (D.S.I.R.), and took charge of the Chemical Research Laboratory at Teddington. His manifold chemical interests and energy suited him admirably for this post; the variety of work was great, so was the output of research. Low-temperature tars, reactions under high pressure, chemotherapy, synthetic resins are only some of the subjects which were dealt with in the laboratory. In one of his presidential addresses to the Chemical Society, he gave an account of some of the chief lines of research in which the laboratory had been engaged, and, incidentally, his hearers learnt many facts of chemical value. This presidential address was of outstanding interest.

Besides work of obviously practical application, research on the purely scientific side of the many problems involved was continued, and a glance through the indexes of *British Chemical Abstracts* gives some idea of Morgan's many-sided activities. He also found time to contribute articles to *Thorpe's Dictionary of Applied Chemistry*, and he edited the chemistry section of the *Encyclopaedia Britannica* (1929), besides publishing books on *Organic Compounds of Arsenic and Antimony* (1918), and *Inorganic Chemistry* (with Burstall, 1936).

On retiring from Teddington, where he is succeeded by a former co-worker, Dr. G. S. Whitby, he became Research Director of the Institute of Brewing; this post he held until his death.

From 1910 to 1912 Morgan was Hon. Secretary of the Chemical Society, and President from 1931 to 1932. He was also President of the Society of Chemical Industry in 1931-1932, and received the Society's medal in 1939; for some years he was Chairman of the Bureau of Chemical Abstracts, and was so at his death.

He graduated as D.Sc. (Lond.) and held various honorary degrees; Sc.D. (Dubl.), LL.D. (Birm., St. And.), M.Sc. (Birm.), and he became F.R.S. in 1915. In addition to his Honorary Membership of our Society, he was a Corresponding Member of the Royal Dublin Society, Honorary Associate of the Manchester College of Technology, F.I.C. and F.C.G.I. In recognition of his war work he was made O.B.E. in 1920, and this was followed by a knighthood in 1936.

Morgan will be sadly missed by his many friends, who remember his kindly acts and recall a humorous and lively companion. Those who did not know him so well personally need only talk to his former students and co-workers (many of whom are already well known in the chemical world) to realise the affection and esteem in which he was held.

J. T. HEWITT

JOHN WHITE, F.I.C.

THE death of John White, on March 30th, in his seventy-seventh year, makes another gap in the now very thin rank of those whose membership of the Society dates back to last century.

White was born at West Bromwich and, after studying chemistry for three years at the Birmingham and Midland Institute, he became a pupil and, later, chief assistant in the laboratory of the late Dr. Bostock Hill. During this time he was appointed Public Analyst for his native town of West Bromwich, carrying on his work in Hill's laboratory until, in 1894, he was appointed "whole time" Public Analyst for the County of Derby, and carried on the duties of this appointment until his retirement in 1933. He was also Public Analyst for the County Borough of Derby, and the Borough of Glossop, Official Agricultural Analyst and Water Examiner for the County and Borough of Derby, Consulting Chemist to the Glossop Sewage Works and to the Derbyshire Agricultural Society, and Gas Examiner for most of the Boroughs and Urban and Rural District Councils within the county.

He was elected a member of this Society in 1893, served as ordinary member of the Council during various periods ranging from 1898 to 1926, and also acted as

Vice-President in 1907-8, and again in 1927-28. He became an Associate of the Institute of Chemistry in 1888 and a Fellow in 1891, and served on its Council in 1910-13. He was for some time Chairman of the Nottingham Section of the Society of Chemical Industry.

White, to those who, like the writer of these notes, knew him intimately, was a man of endearing personality. Under a modest and retiring exterior, he had a wide knowledge of general science, with a supreme sense of public duty and a corresponding zeal for carrying it out with the wise caution of a well-balanced mind. During his earlier days he rarely missed a meeting of our Society, and was always helpful in discussion, although he did not contribute many original papers to our proceedings. Among the few was an interesting paper on "The Occurrence of Barium Compounds in Artesian Well Waters" (ANALYST, 1899, 24, 67); others were on the "Use of Maize as an Adulterant of Oatmeal" (ANALYST, 1895, 20, 30), on "Caper Tea" (ANALYST, 1899, 24, 117), and on "Spurious Cream of Tartar" (ANALYST, 1902, 27, 118).

The writer has pleasant recollections of a Summer Meeting of the Society at Derby during his own Presidency in 1897, arranged by White, aided by our late President, Leonard Archbutt (Chemist to the Midland Railway), on which occasion we were shown over the laboratory and locomotive works of the Railway, the printing and lithographic works of Messrs. Bemrose & Sons, and the works of the Crown Derby Porcelain Manufactory. Among the few survivors of the happy party assembled on that occasion is our old member A. H. M. Muter, still happily active, whose membership dates from the same year (1893) as that of White.

The hobby in which White found change and relaxation from his strenuous work was golf, in which sport he was a creditable performer. For many years he and Archbutt played regularly together on their home links, and the writer well remembers occasional days on links nearer to London, when White joined in "foursomes" with him and Bevan and Clowes or Oscar Guttmann—days stolen by White on the morrow of Society meetings.

On his retirement from office White went to live at Weston-super-Mare, but moved later to Sutton Coldfield, where he died. He was buried at Derby, his funeral service being conducted by the Ven. Dr. Noakes, Archdeacon of Derby, to whom at one time he had been churchwarden at St. John's Church, Derby.

He leaves a widow, a son and two daughters, one of whom is the wife of our member Mr. Stanley Dixon, Public Analyst for Cardiff. BERNARD DYER

Biological Assay of Vitamin D₃

I. Assay Methods at Present in Use, with Particular Reference to Olsson's Radiographic Technique

By A. Z. BAKER, B.Sc., M.R.C.S., L.R.C.P., AND M. D. WRIGHT, B.A., M.B., B.S.

EVER since the discovery^{1,2} of the nutritional importance of animal fats containing fat-soluble vitamin D for the normal calcification of bone, methods of measuring the extent of calcification have been made the basis for estimating the amount of the antirachitic vitamin present in cod-liver oils, etc.

Both the rat and the chick have been extensively used as the test animal for this purpose. Work on chicks was at first hindered by the practical difficulties associated with rearing day-old birds under laboratory conditions. Most of the chick studies are by American workers, a large number of whom have approached

the question of calcification in the bird chiefly from the standpoint of sound practice in poultry rearing. So far, the best-developed method has been the use of the percentage ash-content of the tibia determined under standard conditions.³ It is well recognised that experimental rickets in the bird, unlike its analogue in rats, can be produced without gross disturbance of the mineral balance of the ration, although favourable Ca:P ratios in the diet exert a sparing effect on the vitamin D needed for calcification to proceed normally. A considerable body of information has been accumulated about the influence of other dietary constituents upon the chick's requirement of the calcifying vitamin, the cereal,⁴ and especially the mineral composition of the ration, having been subjected to much study.

In this country the rat has been the experimental subject for vitamin D assay almost to the exclusion of the chick. All the methods in use, however, measure the extent of bone development, with the proviso that experimental rickets in rats demands for its appearance a highly abnormal mineral balance in the diet. The three methods used hitherto for the study of bone structure are (a) McCollum's line test⁵ based on the cure of rickets; (b) estimation of the ash-content of the bones⁶ of the hind limbs of rats maintained on prophylactic levels of vitamin D; (c) radiographic examination of rats undergoing either curative or prophylactic tests.⁷ In addition, a method has been described based upon the increase of weight observed in rats on a supplement of irradiated ergosterol,⁸ but the method, though an actual measure of the vitamin activity, did not show sufficient accuracy to justify its further use.

Much of the earlier work on the chick was concerned with finding the percentage of cod-liver oil which would afford protection from rickets under any given conditions of diet, but the discovery in 1926^{9,10} that vitamin D can be produced by irradiation of ergosterol led to the establishment of irradiated ergosterol as the standard substance for vitamin D assays. The unit of activity was taken as 1 mg. of a standard solution of the material in olive oil, and this unit was adopted by the Pharmaceutical Society in 1927, the Medical Research Council in 1930, and the International Vitamin Conferences of 1931 and 1934.

With the adoption of a vitamin D standard, progress increased. Many of the difficulties inherent in the interpretation of results from animal observations made without reference to a standard substance were controlled and technique was improved. Routine assays of cod-liver oil quickly came to be regarded as essential for examining oils bought and sold with a vitamin D guarantee. Rat methods remained in almost exclusive use in this country, but most American stations used the A.O.A.C. method of comparing oils for agricultural feeding with a standard cod-liver oil.

Anomalous differences in the behaviour of rats and chicks soon became apparent. Rickets appeared when trials were made to protect chicks by feeding to them an amount of irradiated ergosterol equivalent in "rat units" to an amount of cod-liver oil that would have afforded ample protection. In 1928,¹¹ it was stated that 0.0001 mg. of irradiated ergosterol daily would cure rickets in the rat, but that the chick needed as much as 0.01 mg., and that its immunity from overdose of ergosterol was exceedingly high. In 1930, Massengale and Nussmeier¹² studied

the chick requirement of irradiated ergosterol by two methods:—determining the calcium and phosphorus in the serum and the bone ash in the femur. They found that approximately 100 times the number of rat units must be fed as the irradiated product to give a mean value for the percentage of ash in groups of 10 birds as high as was given by 2 per cent. of cod-liver oil in the basal ration. This high requirement of the chick for irradiated ergosterol was widely confirmed in the next few years. Steenbock and his co-workers in 1932¹³ quoted it as between 40 and 120 times the number of rat units that would be sufficient as cod-liver oil, and came to the conclusion that the vitamin D produced by ordinary irradiation of ergosterol is a different substance from the antirachitic factor in cod-liver oil.

The ultimate discovery of vitamin D₃ was approached by deduction from the observation made by several investigators that, although irradiated ergosterol was so inactive in the chick, exposure of the bird to very short periods of irradiation was highly protective.^{14,15,16} Waddell (1934)¹⁷ therefore irradiated an animal extract composed of crude cholesterol, lecithin and fat and found that it was quite as effective for chicks as cod-liver oil, fed in equivalent "rat units." Since then, vitamin D₃ obtained by irradiating 7-dehydrocholesterol has been found to give the same result; it has been isolated, and its constitution has been established.

The calcifying vitamin effective for birds is thus available in pure form as a standard against which to assay oils for poultry feeding. No such standard has yet come into use, and the custom of expressing the vitamin D potency of oils for poultry feeding in International Units as a result of rat assay, using as the standard the official solution of irradiated ergosterol, is still widespread. It should be understood that this test measures the calciferol or vitamin D₂ activity and does not necessarily indicate an equal potency of vitamin D₃ in an oil. The vitamin D₂-like activity of substances is not a reliable indication of their vitamin D₃-like activity, that is, of their efficiency for the chick.

Olsson, in 1936,¹⁸ reported that the amount of vitamin D₃ in chick rations could be related, within certain limits, to the width of the tarso-metatarsal (TMT) distance measured in radiographs of the growing bones (Fig. 1). He considered this a sensitive and satisfactory assay method (Fig. 2, p. 329). Work to confirm and expand his findings has been in progress for the last 18 months in these laboratories; in the earlier tests a provisional standard reference oil, BZ1, which had been used in an extensive series of tests on rats, and was kindly provided by British Colloids, Ltd., was employed. This oil was suitably stored and fed in graded amounts in a series of chick tests, with reasonably uniform results. More recently it has been possible to obtain, through the British Standards Institution (Vitamin D Panel of Sub-committee C 11/2: Chick Test for Veterinary Cod-liver Oil) supplies of pure vitamin D₃ in solution for use as a standard for direct comparison.

Olsson's finding, that the width of the TMT distance can be used as a basis of vitamin D₃ assay, has been confirmed but, following a suggestion based on statistical analyses by Mr. E. C. Fieller, it has been found that the relation is more accurately expressed by comparing the logarithm of the TMT distance,

Fig. 1



(a)

(a) Normal



(b)

Tarso-metatarsal distance
(b) Slight degree of rickets



(c)

(c) Severe degree of rickets

rather than the distance itself, with the logarithm of the dose. The first table below, however, is based on Olsson's method of expression.

The principles of Olsson's radiographic method have been used in our tests, but with slight differences in some particulars, which will be described. Olsson at first recommended 6 weeks as the optimum age for radiographic examination, but in a later paper¹⁹ he suggests 4 to 6 weeks. We have taken X-ray photographs at 4, 5 and 6 weeks and have recorded most of the results at 5 weeks. This period

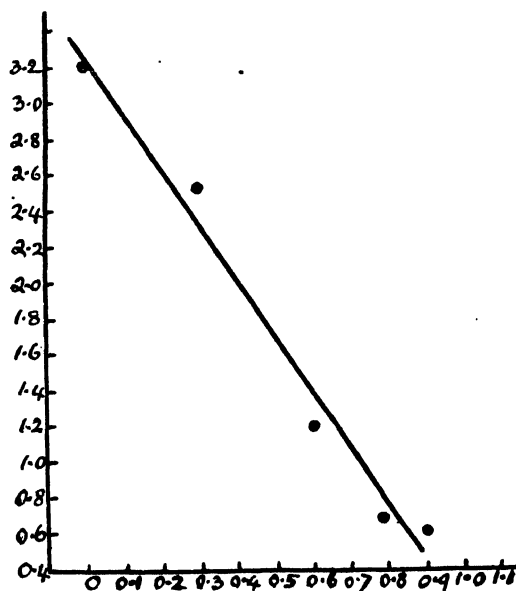


Fig. 2

was chosen in preference to 4 weeks, as the slightly heavier bones yielded radiographs rather more easily measured than those obtained at 4 weeks of age. The possible increase in accuracy by using data obtained after 5 weeks' feeding, following a one-week depletion period, is under investigation.

TABLE I

| Level of oil fed Per Cent. | Mean TMT | | Standard deviation | |
|-------------------------------|----------------|----------------|--------------------|---------|
| | 4 weeks mm. | 6 weeks mm. | 4 weeks | 6 weeks |
| 0.50 | 1.35 | 1.27 | 0.164 | 0.159 |
| 0.25 | 1.37 | 1.31 | 0.131 | 0.181 |
| 0.125 | 1.50 | 1.43 | 0.111 | 0.185 |
| 0.0625 | 1.88 | 1.74 | 0.596 | 0.485 |

TECHNIQUE.—Chicks Used.—These are sex-linked cockerels (Light Sussex and Rhode Island Red), obtained as day-old chicks from a known reliable source.

Method of Housing.—The chicks are reared, 20 in a compartment (dimensions 10" × 11" × 66") in Summit Morcote all-metal brooders specially modified by Messrs. Cope & Cope, Reading, for our purpose. Each brooder contains 6 units in

three rows of two and each unit contains a warm chamber and an unheated run. The floors are of wire mesh over metal trays. Heating is by electricity, the element being placed in the roof of the closed portion and the temperature thermostatically controlled. The battery of brooders is housed in a brick building, from which daylight is entirely excluded by insulated shutters, light being supplied during 12 hours a day by electric lights so placed as to give even illumination to all the compartments. Each unit is provided with a trough containing water kept at a constant level; this prevents crowding.

Ventilation is effected by baffled inlets near floor level, the outlets being along a central ceiling shaft communicating with an exhaust fan of controlled speed.

Food.—The chicks are all given a basal ration to which are added different quantities of the test oil in olive oil, to make a total addition of 1 per cent. of oil. The *basal* ration is as follows:—maize meal, 35; middlings, 24; bran, 16; dried skimmed milk, 8; meat meal (albumenoids 50 per cent.), 10; dried grass, 5; salt, 0.5; ground limestone, 1.5; olive oil, 1.0 per cent. The grass meal was included for its high carotene content; its use made it unnecessary otherwise to modify the vitamin A contents of the different diets. *Analysis.*—protein, 18.19; fat, 5.27; ash, 6.85; ash, calcium, 1.82; phosphorus, 0.98; fibre, 4.7 per cent. $\text{Ca/P} = 1.85$; vitamin A and carotene, 210 I.U. per 100 g.

The ingredients are weighed on a balance recording down to 0.25 oz. and mixed in an electric mixer for 20 minutes. A portion is then removed, the oil is mixed with it by hand and the food is sieved, after which it is returned to the bulk and mixed for a further 20 minutes. Determinations of oil in random samples of mash prepared in this way have shown that such mixtures are uniform.

Depletion Period.—In recent tests the birds have been reared for the first week on the basal ration without vitamin D supplement in order to exhaust their yolk sac store and give the greatest possible uniformity among those selected for the test.

Treatment of the Test Oils.—The uniform admixture of the test oils and olive oil is also effected by electric stirring in a Hobart machine. The basal diet used in conjunction with an adequate supply of cod-liver oil produces sturdy well-feathered chicks, weighing approximately 10 oz. at 4 weeks and 17 to 20 oz. at 6 weeks.

Procedure.—At first 40 chicks were fed at each level of any test oil, usually in 2 units of 20 each. A number of oils are fed simultaneously, and the groups are so arranged as to reduce as far as possible differences which might arise from different positions in the house. More recently the allocation of groups of 20 birds at four or more levels of the standard and of the test oil respectively has made it possible to assay with considerable accuracy, and with economy of time, samples of widely differing vitamin activity. The value of using several levels under these conditions is stressed, for this provides a good "spread" from which to calculate the slope of the curve of response.

X-ray Technique and Records.—The tarso-metatarsal joints are photographed in an anteroposterior position, an exposure of 0.25 second at 12 inches, 12 ma. Position 2 Kv (Victor X-ray apparatus) being given. The tarso-metatarsal distance is measured on the film (supported on a viewing screen) by a vernier microscope magnifying 5 diameters (Baker) and reading to 0.02 mm. Olsson's

method of magnifying 8 to 12 diameters has not been found possible on the films available in this country, owing to the size of grain.

Accuracy has been increased by the following slight modification of Olsson's technique, suggested by Mr. E. T. Halnan of the School of Agriculture, Cambridge. A fine line is drawn with a razor blade tangential to the proximal end of the fused metatarsus, and the distance is taken between this line and the nearest point of the central tarsal bone. By this modification the range of one group of figures was reduced from 0.62–1.56 to 0.78–1.02 mm., and that of another from 1.34–4.88 to 1.52–4.52 mm.

Results.—The method has been used with several different samples of cod-liver oil, with various fish oils of high potency and with cod-liver oil concentrates, and we are able to confirm Olsson's finding that it is accurate and economical. The modification of using log TMT, rather than the distance itself, was introduced to equalise the variability of responses to different doses.

TABLE II

| | Test dose per 30 g. of mash cg. | No. of chicks | Log of mean response, mm. \times 10 | σ | Coefficient of variation |
|-----|--|------------------|---|----------|--------------------------------|
| I | 4 | 17 | 0.938 | 0.120 | 12.8 |
| II | 3 | 18 | 1.132 | 0.163 | 14.4 |
| III | 2 | 16 | 1.252 | 0.177 | 14.1 |
| IV | 1 | 16 | 1.396 | 0.163 | 17.7 |

On the scale on which the work is done in these laboratories it has been found possible, after practice, to evaluate the results graphically to a figure within 5 to 8 per cent. of that obtained by full statistical analysis; however, this has been carried out in every test to obtain the error of each assay and to detect and assess variations from linearity.

Statistical analysis is of rather greater importance when the scale of the work is more limited than is described in this paper, and the additional information thereby obtained will generally amply repay the labour involved. The method used for analysis is set out in the appendix.

Many oils have been examined in these laboratories by the radiographic method, and the following are typical of the results obtained:

TABLE III

| Cod-liver oil | Value u.p.g. | No. of chicks | Percentage limits of error | | |
|------------------|-----------------|------------------|----------------------------|------------|--|
| | | | P = 0.95 | P = 0.99 | |
| Sample A | 65 | 81 | 80–126 | 74–135 | Standard reference oil, C.L.O., taken as 200 units per g. (assayed on rats) |
| B | 82 | 60 | 74–135 | 68–148 | |
| C | 131 | 63 | 84–119 | 80–126 | |
| D | 100 | 80 | 84–119 | 79–126 | |
| E | 70 | 70 | 84–119 | 79–126 | |
| F | 89 | 84 | 83–121 | 78–129 | |
| G | 111 | 76 | 86–117 | 81–123 | |
| H | 97 | 76 | 85–118 | 81–124 | |
| I | 46 | 67 | 80.6–124 | 75.5–132.5 | |
| | | | | | Pure vitamin D ₃ |

Samples of other liver oils at suitable dilutions have also been examined with comparable results.

TABLE IV

| High potency oils | Value u.p.g. | No. of chicks | Percentage limits of error | | |
|-------------------|--------------|---------------|----------------------------|----------|--------------------------------|
| | | | P = 0.95 | P = 0.99 | |
| Sample A | 35,000 | 114 | 85-117 | 81-123 | Standard reference oil, C.L.O. |
| B | 3,400 | 65 | 82-122 | 77-130 | " " " |
| C | 5,500 | 76 | 74-135 | 68-148 | " " " |
| D | 14,300 | 54 | 90-111 | 87-115 | " " " |
| E | 11,080 | 49 | 80.6-124 | 75.5-132 | Pure vitamin D ₃ " |

TABLE V

| Method | Standard deviation | Slope | No. of chicks receiving any one oil | Percentage limits of error | |
|---------------------|--------------------|-------|-------------------------------------|----------------------------|----------|
| | | | | P = 0.95 | P = 0.99 |
| Radiographic | 0.293 | -1.29 | 200 | 93-107 | 91-110 |
| | | | 100 | 90-111 | 88-114 |
| | | | 50 | 86-116 | 83-121 |
| | | | 20 | 79-126 | 74-135 |
| | | | 10 | 72-138 | 65-153 |
| Bone ash, per cent. | 2.364 | 8.672 | 200 | 92-109 | 89-112 |
| | | | 100 | 88-113 | 85-118 |
| | | | 50 | 84-119 | 79-126 |
| | | | 20 | 76-132 | 69-144 |
| | | | 10 | 67-148 | 60-167 |

Some comparisons have been made between the error given by the radiographic chick method and that obtained with birds on the same doses of the oils under examination, tested by Dr. Magnus Pyke of these laboratories, under the standard conditions laid down by the Association of Official Agricultural Chemists (U.S.A.), but with a separate determination of the ash percentage of each bone. Figures from such a test are given in Table V, from which it is clear that experimental error is of the same order with Olsson's technique as in the A.O.A.C. method. The radiographic method avoids certain disadvantages inseparable from the A.O.A.C. technique, *viz.*: (1) The necessity for killing the birds in order to collect the experimental data. (2) The long time consumed in dissecting the bones and the difficulty of reducing this beyond certain limits, for the accuracy of the results depends to a considerable extent upon skilful and uniform dissection of all the samples. Unless individual bones are separately ashed, the error of any experiment cannot be stated. Group ashing has been widely practised in assays by the A.O.A.C. technique.

The main practical advantages of the radiographic technique are: (1) That a permanent record of assays can be kept for reference at any future date. The X-ray photographs can be measured by different workers, if desired, and allowance made for any individual differences. (2) That the chicks are rearable after the experiment, so that larger numbers can be used without a corresponding increase in expense. Greater accuracy is thus obtained at lower cost than in the A.O.A.C. method. In this laboratory 840 chicks are used at one time for a series of tests and are later reared under farm conditions for the table.

SUMMARY.—1. Since normal osteogenesis in birds is promoted far more economically by vitamin D₃ than by calciferol, stress is laid on the practical importance of basing assays of the antirachitic value of liver oils for poultry feeding upon bird tests, using pure vitamin D₃ as the standard of reference.

2. Olsson's technique has been investigated and modified. Confirmation has been obtained of his claim that the TMT distance in chicks is a function of the logarithm of the amount of vitamin D₃ supplied in the diet, but it is found that using the logarithm of the TMT gives greater accuracy than using the actual distance.

3. The error of the Olsson method has been estimated in a number of assays and found to be approximately the same as that of the A.O.A.C. method conducted in the same laboratory.

4. The Olsson method has been found to be of relatively low cost in laboratories where a suitable X-ray plant is available.

Our thanks are due to Mr. E. C. Fieller, for the statistical treatment of assays and for his helpful criticism and advice; also to Mr. L. Jones, of British Colloids, Ltd., for duplicating the ash determinations.

APPENDIX.—This appendix illustrates the method of statistical analysis used in the vitamin D assays of oils. In a typical assay readings were obtained from groups of 17, 18, 18, 19 and 18 chicks, which had received supplements of 5, 10, 15, 20 and 30 units of standard oil per 100 g. of food respectively. Table A, column (2) gives the logarithm of the dose in units/20 g. of food, and column (3) the number of readings per group. Column (5) gives the sum of the responses, which are recorded as $[1 + \log_{10} (\text{TMT distance in mm.})]$, in the successive groups, and column (10) the sum of their squares.

TABLE A

| Dose (1) u/100 g. | (2) x_1 | (3) n_1 | (4) $n_1 x_1$ | (5) $n_1 y_1$ | (6) y_1 | (7) $n_1 x_1 y_1$ | (8) $n_1 x_1^2$ | (9) $n_1 y_1^2$ | (10) Σy_1^2 | (11) $f_1 s_1^2$ | (12) f_1 | (13) s_1^2 |
|----------------------|--------------|--------------|------------------|------------------|--------------|----------------------|--------------------|--------------------|------------------------|---------------------|---------------|-----------------|
| 5 | 0.0000 | 17 | 0.0000 | 24.33 | 1.431 | 0.000000 | 0.000000 | 34.8205 | 35.2185 | 0.3980 | 16 | 0.02487 |
| 10 | 0.3010 | 18 | 5.4180 | 21.76 | 1.209 | 6.549760 | 1.630818 | 26.3054 | 25.6660 | 0.3606 | 17 | 0.02121 |
| 15 | 0.4771 | 16 | 7.6336 | 16.84 | 1.052 | 8.034364 | 3.641991 | 17.7241 | 18.3076 | 0.5835 | 15 | 0.03890 |
| 20 | 0.6021 | 19 | 11.4399 | 19.19 | 1.010 | 11.554399 | 6.887964 | 19.3819 | 19.9687 | 0.5868 | 18 | 0.03260 |
| 30 | 0.7782 | 18 | 14.0076 | 16.94 | 0.941 | 13.182708 | 10.900714 | 15.9424 | 16.3296 | 0.3872 | 17 | 0.02278 |
| Sums (Σ) | | 88 | 38.4991 | 99.06 | | 39.321131 | 23.061487 | 114.1743 | 116.4904 | 2.3161 | 83 | |
| | | | Correction terms | .. | | 43.337737 | 16.842963 | 111.5100 | | | | |
| | | | Differences | .. | .. | -4.016606 | 6.218524 | 2.6643 | | | | |

Column (6) contains the mean responses, and column (9) their products by the group sum. By subtracting this from the entry in column (10), we obtain the sum of the squares of the deviations of the individual responses in each dosage group from their mean (column 11). Column (12) gives the corresponding degrees of freedom (group frequency less one); dividing this into the entry in column (11) we obtain the estimated variance of individual responses to the successive doses (13). These variances, and those for the dosage groups on the test oil, prove to be in good agreement with each other, and we are therefore justified in pooling them and in evaluating the assay by the method described by Irwin.²⁰

The calculations for the slope of the log (dose)-response line are contained in columns (7) and (8), which are explained by their headings; the correction terms subtracted from their totals are obtained from the totals of columns (3), (4) and (5) as $(\Sigma n x) (\Sigma n y) / (\Sigma n)$ and $(\Sigma n x)^2 / (\Sigma n)$. For later use we also subtract a term $(\Sigma n y)^2 / (\Sigma n)$ from the total of column (9).

The test oil was fed at levels of 1, 2, 3 and 4 cg./32 g. of food, and similar calculations to those in Table A lead to the totals given in Table B (x_1 = log. of dose in cg./32 g. of food).

TABLE B

| | n_i | $n_i x_i$ | $n_i y_i$ | $n_i x_i y_i$ | $n_i x_i^2$ | $n_i y_i^2$ | Σy_i^2 | $f_i s_i^2$ | f_i |
|-------------------|-------|-----------|-----------|---------------|-------------|-------------|----------------|-------------|-------|
| Sums (Σ) | 67 | 23.6395 | 79.68 | 25.345031 | 11.709770 | 94.2655 | 95.8172 | 1.5517 | 63 |
| Correction terms | | .. | .. | 27.760534 | 8.340686 | 92.3962 | | | |
| Differences | .. | .. | .. | -2.415503 | 3.369084 | 1.8693 | | | |

We now calculate the means

$$\bar{x}_1 = 38.4991/88 = 0.4375$$

$$\bar{y}_1 = 99.06/88 = 1.1257$$

$$\bar{x}_2 = 23.6395/67 = 0.3528$$

$$\bar{y}_2 = 79.68/67 = 1.1743$$

and the slope

$$b = -\frac{4.0166 + 2.4155}{6.2185 + 3.3691} = -\frac{6.4321}{9.5876} = -0.6709$$

The log. (potency ratio) is estimated to be

$$M = (\bar{x}_1 - \bar{x}_2) - (\bar{y}_1 - \bar{y}_2)/b = 0.012151 = \log. (1.0285).$$

We therefore estimate that 31.25 mg. of the test oil contain $5 \times 1.0285 = 5.143$ units of D₃, i.e. that its potency is 164.6 u/g. To verify that the series of points (x, y) do not deviate significantly from straight lines, we calculate the sum of squares (for 6 degrees of freedom)

$$2.6643 + 1.8693 - \frac{(6.4321)^2}{9.5876} = 0.2184$$

and compare the corresponding mean square, 0.0364, with the pooled group variance (with 146 degrees of freedom)

$$s^2 = (2.3161 + 1.5517)/(83 + 63) = 0.02649.$$

Since s^2 is not significantly smaller than the mean square 0.0364, the linearity of the log (dose) response relation is verified, and we can calculate the approximate error of M from Irwin's formula:

$$\sigma_M^2 = \frac{s^2}{b^2} \left\{ \frac{1}{\Sigma n_1} + \frac{1}{\Sigma n_2} + \frac{(\bar{y}_1 - \bar{y}_2)^2}{b^2 \{ \Sigma n_1 (x_1 - \bar{x}_1)^2 + \Sigma n_2 (x_2 - \bar{x}_2)^2 \}} \right\}$$

$$= \frac{0.026492}{0.450076} (0.011364 + 0.014925 + 0.000540) = 0.0015797$$

Whence $\sigma_M = 0.03975$.

For 146 degrees of freedom, the 5 per cent. and 1 per cent. levels of Student's t are 1.977 and 2.610. Since $1.977 \sigma_M = 0.0786$, $2.610 \sigma_M = 0.1037$, the limits of M are

$$\text{for } P = 0.95 : 0.0122 \pm 0.0786, \text{ i.e. } \bar{1}.9336 \text{ and } 0.0908,$$

$$\text{for } P = 0.99 : 0.0122 \pm 0.1037, \text{ i.e. } \bar{1}.9085 \text{ and } 0.1159.$$

The limits for the D₃ content of 31.25 mg. of test oil are therefore

$$\text{for } P = 0.95 : 5(0.858) \text{ units to } 5(1.233) \text{ units,}$$

$$\text{for } P = 0.99 : 5(0.810) \text{ units to } 5(1.306) \text{ units.}$$

and those for the content of 1 g. of test oil

$$\text{for } P = 0.95 : 137 \text{ units to } 197 \text{ units,}$$

$$\text{for } P = 0.99 : 130 \text{ units to } 209 \text{ units.}$$

REFERENCES

1. E. Mellanby, *J. Physiol.*, 1918, **52**, XL.
2. V. Korenchevsky, 1922, *Special Report Series*, Medical Research Council, London, No. 71.
3. A.O.A.C. *Methods*, 1937, **20**, 1, 72.
4. H. D. Branton, *Poultry Science*, 1932, **12**, 335.
5. E. V. McCollum, N. Simmonds, P. G. Shipley and E. A. Parks, *J. Biol. Chem.*, 1922, **51**, 41.
6. H. Steenbock and A. Black, *J. Biol. Chem.*, 1924, **61**, 405.
7. R. B. Bourdillon, H. M. Bruce, C. Fischmann and T. A. Webster, 1931, *Special Report Series*, Medical Research Council, London, No. 158.
8. K. H. Coward, K. M. Key and G. E. Morgan, *Biochem. J.*, 1932, **26**, 1585.
9. O. Rosenheim and T. A. Webster, *id.*, 1927, **21**, 389.
10. A. Windaus and A. F. Hess, *Nach. Ges. Wiss., Göttingen*, 1926, **2**, 175.
11. H. Kreitmair and T. Moll, *Munch. Med. Woch.*, 1928, **75**, 637.

12. O. N. Massengale and M. Nussmeier, *J. Biol. Chem.*, 1930, **87**, 423.
13. H. Steenbock, S. W. F. Kletzien and J. G. Halpin, *id.*, 1932, **97**, 249.
14. H. T. Scott, E. B. Hart and J. G. Halpin, *Poultry Science*, 1929, **9**, 65.
15. W. C. Russell, O. N. Massengale and C. H. Howard, *J. Biol. Chem.*, 1928, **80**, 155.
16. W. C. Russell and C. H. Howard, *id.*, 1931, **91**, 493.
17. J. Waddell, *id.*, 1934, **105**, 711.
18. N. Olsson, *Arch. fur. Geflügelkunde*, 1936, **10**, 11-12.
19. ——— *Kungl. Fyriografiska Sällskapet i Lund Forhandlingar*, 1936, **9**, 1.
20. J. C. Irwin, *Statistical Method Applied to Biological Assays*, Supp. to *Trans. R.S.S.*, 1937, **IV**, 1-48.

THE RESEARCH LABORATORIES

VITAMINS LIMITED

23, UPPER MALL, HAMMERSMITH, W.6

January, 1940

A New Group Separation for the Quantitative and Qualitative Analysis of Phosphates

BY G. J. AUSTIN

HITHERTO no method of analysis suitable for the quantitative separation of the common metals has been applicable in presence of more phosphate than is equivalent to the amount of the iron group metals present. The following method is intended for such instances and aims at placing the analysis of phosphates on a level with ordinary analysis in both accuracy and simplicity.

Smith¹ found that a qualitative separation of the iron group metals in presence of excess of phosphate is possible at pH 3. Quantitatively, the optimum pH has been found to be 3.2-3.4. Haring and Leatherman² have shown that cobalt sulphide can be quantitatively precipitated at pH 4. Haring and Westfall³ have shown that nickel sulphide is precipitated at pH 4.5. Similarly, Fales and Ware⁴ have shown that zinc sulphide is completely precipitated at pH 2.0 and above. For the rapid precipitation of all three sulphides, pH 4.6-4.8 has been found best. The remaining principles of this new method are frequently employed in analysis.

ADVANTAGES OF THE METHOD.—Phosphate, instead of being removed, is employed to advantage. The adsorption losses and additional filtrations inevitably accompanying the removal of the phosphates are thus avoided. The method can be applied with facility when the phosphate is present in large excess over the iron and aluminium, under which conditions the classical methods fail.⁵

The metals barium and strontium, which are especially liable to be carried down by the iron group precipitate when phosphate is present, are removed first. The risk of losing these metals, owing to sulphate being present in the reagents used, is thus avoided and their detection is aided. There is no tendency for precipitates of a colloidal nature to be formed; zinc sulphide and calcium oxalate are often troublesome when precipitated from ammoniacal solutions. The iron

AUSTIN: A NEW GROUP SEPARATION FOR THE QUANTITATIVE AND

group phosphates can be filtered off much more readily than the basic acetates. Very selective reactions are employed and all the chemicals used are common and obtainable very pure. Adsorption errors compare very favourably with those in the classical methods of analysis.

PRINCIPLES OF THE METHOD.—(1) Groups 1 and 2 are removed as usual.

(2) Barium and strontium are precipitated as sulphates from a solution approx. $N/5$ in hydrochloric acid and of about 25 ml. in volume.

(3) Iron, aluminium and chromium are precipitated as phosphates at pH 3.2 to 3.4.

(4) The solution is then buffered to pH 4.6–4.8 and treated with hydrogen sulphide at 70° to 80° C. Zinc, nickel and cobalt sulphides are precipitated.

(5) After most of the hydrogen sulphide has been boiled off the manganese is precipitated as dioxide by oxidising with sodium hypochlorite.

(6) The calcium is precipitated as oxalate.

(7) The solution is finally made ammoniacal and magnesium is precipitated as phosphate.

(8) No provision is made for the detection of sodium and potassium, which must be tested for separately; this is more convenient and reliable, because a direct method minimises contamination with sodium and potassium from glassware and reagents and does not introduce large quantities of ammonium salts.

REAGENTS.—The following reagents are required:

5 N hydrochloric acid (dilute the concentrated acid to sp.gr. 1.084).

2 N sodium hydroxide solution.

M potassium dihydrogen phosphate solution (13.7 g. per 100 ml.).

3 N and N sodium acetate solutions.

M citric acid (21 g. per 100 ml.).

Sodium hypochlorite solution (commercial solution diluted to contain about 5 per cent. of available chlorine).

Bromophenol blue (0.1 g. of solid ground in a mortar with 4.1 ml. of $N/20$ sodium hydroxide solution and diluted to 250 ml.

$N/20$ sulphuric acid; $N/10$ and $N/100$ acetic acid; $N/10$ nitric acid.

3 N sodium nitrite solution (20 g. per 100 ml.).

A NEW GROUP SEPARATION FOR THE QUALITATIVE AND QUANTITATIVE ANALYSIS OF PHOSPHATES

To cold soln. of 0.5 g. in 20 ml. add 2 *N* NaOH, 2-3 drops at a time and with stirring until a slight ppt. persists without appreciably dissolving in 20 secs. Add 1 ml. of 5 *N* HCl. Heat to incipient boiling, add 2 ml. of 10 per cent. Na₂SO₄ and, if there is a ppt. add another 2 ml. of Na₂SO₄ and heat in a boiling water-bath for 15 mins. Filter and wash with *N*/20 H₂SO₄.

Ppt. **Solution.** Dilute to 100 ml. and while heating add 3 ml. of glacial acetic acid and then 2 ml. of 3 *N* sodium nitrite. Boil $\frac{1}{2}$ min. Add 0.4 ml. of *M* KH₂PO₄ to ensure enough phosphate present to form Fe, Al and Cr phosphates. Add 3 ml. of *N* sodium acetate. Then try spot test with bromophenol blue and, if a yellow colour is obtained, add 1 ml. portions of sodium acetate soln. until a grey-green colour is obtained (pH 3.2 to 3.4). For quantitative analysis spot tests must now be washed back into the soln. Finally boil for a few secs. (3 mins. if Cr is present). Filter through a fast paper and wash ppt. preferably with hot *N*/100 acetic acid.

SrSO₄,
BaSO₄,

Try flame test and/or boil with 50 ml. of *N* Na₂CO₃. Decant through filter. Repeat process. Dissolve carbonates in acetic acid. Add K₂CrO₄; ppt. = BaCrO₄. To soln. add H₂SO₄; ppt. = SrSO₄.

| | | | |
|---|--|--|-----------------------------|
| Ppt. FePO₄, AlPO₄, CrPO₄. | Solution. Heat to boiling and ignore any ppt.; add 15 ml. of 3 <i>N</i> sodium acetate soln. Pass a fairly rapid stream of H ₂ S through the hot soln., preferably at 70° to 80° C. Absence of a ppt. in 2 mins. shows absence of Zn, Co or Ni; otherwise continue passage of gas for 5 to 10 mins. Heat nearly to boiling, filter and wash with <i>N</i> /10 acetic acid. | | |
| Ppt. ZnS CoS NiS or If Zn is present, white ZnS ppts. before the black CoS and NiS. | Solution (150-250 ml.). Boil off H ₂ S. To boiling soln. add NaOCl soln. at rate of about 1 drop per sec. Absence of a brownish ppt. with 2 ml. shows absence of Mn; otherwise continue treatment until ppt. darkens. Filter and wash ppt. once or twice with water, then with <i>N</i> /10 HNO ₃ and finally with water. | | |
| Ppt. MnO₂ To confirm, extract with KHC ₄ O ₆ , deep red soln. proves Mn. | Solution. To hot soln. add 5 ml. of <i>M</i> citric acid and 20 ml. of sat'd. ammonium oxalate soln. After standing at least 5 mins. filter and wash with hot water, preferably containing a few drops of ammonium oxalate. | | |
| Ppt. CaC₂O₄.H₂O | Solution. Concentrate to about 150 ml., add 5 ml. of KH ₂ PO ₄ and to hot soln. add a slight excess of NH ₃ , stir for a few mins. and finally add 20 ml. of conc. NH ₃ . Stand overnight. Filter and wash with 2 per cent. NH ₃ . | | |
| | <table> <tr> <td>Ppt. MgNH₄PO₄.6H₂O</td><td>Solution. Reject.</td></tr> </table> | Ppt. MgNH₄PO₄.6H₂O | Solution. Reject. |
| Ppt. MgNH₄PO₄.6H₂O | Solution. Reject. | | |

NOTES ON THE PROCEDURE.—(1) *Preparation of the Solution.*—The quantity of metals present in the test solution should not be equivalent to more than 0.5 g. of the phosphates, so that if the sample is a metal or is mostly oxides it is advisable to take less of it. The 0.5 g. limit does not include Group 1 and 2 and alkali metals.

Organic substances or much ammonium salt must be absent. Under the conditions employed, ammonium salts prevent the oxidation of Mn to the dioxide;

in the absence of Mn ammonium salts do no harm and may be used in place of the sodium salts specified.

The solution of the substance should preferably be in dilute HCl and contain no metals of Group 1 or 2. Such a solution is obtained after Group 2 treatment, but would probably need concentrating to reduce to 20 ml.; this is essential only when strontium is present. If the solution is very strongly acid it is advisable to remove the excess acid by evaporation. Chromates are best first reduced, but provision is made in the scheme for this as well as for the oxidation of iron.

(2) *Precipitation of Barium and Strontium Sulphates*.—If sulphate is present in the test solution, and therefore barium and strontium are known to be absent, treatment with sodium sulphate may be omitted, and it is possible to proceed directly to the iron group separation.

The barium and strontium sulphates adsorb a small proportion of any ferric iron present, but, as at least 90 per cent. is left in solution, the adsorption of iron may be neglected for qualitative analysis. In quantitative analysis, when much iron is present together with barium or strontium, the adsorption of iron may be made negligible by reducing to the ferrous state as follows:

To the boiling solution in a conical flask add 1-ml. portions of 0.5 *N* hydriodic acid or sodium iodide until no more iodine is liberated; boil down to 20 ml. and proceed. Potassium iodide should not be used, because barium sulphate adsorbs potassium salts, giving high results. It is best to use a 100-ml. beaker when precipitating the sulphates.

If strontium is to be determined, heating in the water-bath should be continued for 30 minutes (or the solution left overnight), but for qualitative analysis 10 to 15 minutes are sufficient. When the substance contains a large proportion of calcium an excess of more than 0.2 g. of sodium sulphate should be avoided, but otherwise the specified 4 ml. of the solution may be used. When much calcium is present, the solution is best maintained at 25 ml. during the heating in the bath, and the sulphates should be washed until the washings give no reaction for calcium. Results are somewhat low for strontium when calcium is present in great excess of the amount of strontium. Traces of strontium that escape precipitation as sulphate are precipitated along with the calcium as oxalate.

(3) *Iron Group Phosphates*.—When it is known that the substance contains sufficient phosphate to form the iron group phosphates, it is undesirable to add more phosphate. The adsorption of metals and of phosphoric anhydride increases with the amount of excess phosphate. The adsorption of metals increases with rise in *pH*, but the adsorption of phosphoric anhydride decreases slightly. Dilution does not reduce the adsorption of zinc, but greatly reduces the adsorption of phosphoric anhydride.

If, after adding the 2 ml. of 3 *N* sodium nitrite solution and boiling, 1 ml. of *M* potassium dihydrogen phosphate solution is added and there is little or no precipitate, very little iron or aluminium can be present; in that event no more potassium dihydrogen phosphate solution need be added. If the solution is very green, indicating the presence of much chromium, or if in doubt, add 4 ml. of potassium dihydrogen phosphate solution. The *pH* at this stage is 2–2.5.

The nitrite assists in obtaining the phosphates in a good condition for filtration,

ensures oxidation of the iron and reduction of chromates, and will decompose the excess of iodide that may have been used to reduce iron; in that event, after addition of the nitrite, boiling should be continued until all the iodine is expelled. It may be necessary to stir the solution while heating, in order to prevent bumping.

Except when chromium or zinc is present, a single precipitation will usually suffice for quantitative analysis. When chromium and zinc are both present a single precipitation suffices for qualitative tests.

When re-precipitation is necessary, the phosphates should be dissolved in the minimum quantity of hydrochloric acid, 0.5 ml. of *M* potassium dihydrogen phosphate solution and 3 ml. of glacial acetic acid added, and the solution diluted to approx. 100 ml., heated to boiling, buffered to *pH* 3.2–3.4 by adding alkali acetate, boiled and filtered. The filtrate should be evaporated and most of the acetic acid expelled before adding it to the main filtrate. Throughout this method it is best to avoid boiling the solution in order to concentrate; it is preferable to evaporate the washings or second filtrate separately before adding them to the main solution.

Except when the solution is highly coloured, the buffering to *pH* 3.2–3.4 can be done with the bromophenol blue as internal indicator; the colour will change from yellow to greyish-green. A spotting method is always applicable and is more accurate, and a spot test should always be made to make certain of the *pH*. If there is any doubt about the correct colour, buffers of *pH* 3.0, 3.2 and 3.4 should be prepared by taking 3 ml. of acetic acid and 1, 2 and 4 ml. of *N* sodium acetate solution, respectively, and diluting to 100 ml. in each instance. These solutions should be used for spot tests; buffering will then cause no difficulty.

The solubility of iron phosphate rises very rapidly below *pH* 2.0 and that of aluminium phosphate below 3.0; when large amounts of both phosphate and zinc are present zinc phosphate begins to separate from boiling solutions at *pH* 3.6, nickel, cobalt and manganese at rather higher *pH* values, and calcium phosphate at 4.5. At *pH* 3.2–3.4 there are left dissolved 0.03 to 0.01 mg. of iron and 0.05 to 0.02 mg. of aluminium. A large excess of phosphate does not reduce the solubility of the iron and aluminium phosphates; *pH* is the primary factor.

Determination of Iron and Aluminium as Phosphates.—There is a tendency for too high results to be obtained owing to adsorbed P_2O_5 , but a slight excess of P_2O_5 is necessary to prevent hydrolysis, otherwise low results are obtained. By adding the phosphate at the stage specified, particularly if the potassium dihydrogen phosphate is added slowly, the adsorption of P_2O_5 is greatly reduced. This is because, after the iron and aluminium phosphates have been precipitated, they adsorb little of the P_2O_5 subsequently added and much less than if all the phosphate has been added before precipitation occurred. Providing adsorption of metals is negligible, fair results can be obtained by weighing amounts up to 50 mg. of iron and aluminium phosphates; also larger amounts, provided that an excess of P_2O_5 was not originally present and the potassium dihydrogen phosphate is added slowly. Results are then usually not more than 3 per cent. high, and can be greatly improved by diluting to 200 ml. instead of 100 ml., as specified in the table.

Results correct to within 1 per cent. for amounts of 0.1 to 0.5 g. of iron and aluminium phosphates, and correct to within 1 mg. for amounts less than 0.1 g., can be obtained by re-precipitating as already described; at the same time adsorption of metals is reduced. The phosphates are washed with hot *N*/100 acetic acid until the washings give practically no reaction with silver nitrate, dried and ignited, gently at first, finally at bright red heat, until constant in weight. Aluminium phosphate, being very hygroscopic, must be weighed rapidly. A mixture of iron and aluminium phosphates can be dissolved in hydrochloric acid, iron determined volumetrically, and the aluminium obtained by difference.

All the alkali salts are removed by washing until free from chlorine. Washing with ammonium acetate or nitrate solution tends to give low results, owing to hydrolysis, whilst washing with ammonium phosphate solution tends to give high results owing to adsorption of P_2O_5 . Ignition with filter-paper tends to reduce the phosphates, and for this reason filter-paper pulp as an aid to filtration is not advisable when the phosphates are to be weighed. Filtration is facilitated by keeping the funnel in a hot-water jacket or by placing it in a hot air-bath.

Chromium.—By this method all but about 2 mg. of chromium is precipitated when iron and aluminium are absent, but in their presence not more than 0.5 mg. of chromium remains in solution, and amounts of less than 2 mg. are almost completely carried down by 10 mg. of aluminium or iron phosphate. Traces of chromium remaining in solution do not interfere with subsequent separations and can be recovered after the magnesium test by boiling with tannic acid at *pH* 6–7.

Failure of chromium to be precipitated from acetate solutions appears to be due to complexes formed with the chloride, acetate and other ions (see Ref. 6). The stability of such complexes apparently increases with rise in *pH* from 2 to 7, for above *pH* 3 the precipitation of chromium phosphate becomes more and more sluggish, and only at *pH* 2.5 to 3.5 is precipitation in presence of chloride and absence of iron and aluminium, reasonably complete. In absence of iron and aluminium, the precipitation of chromium basic acetate fails at any *pH*.

The method can be used qualitatively when chromium is present and aluminium absent, but satisfactory quantitative separations can only be obtained when the chromium is present together with at least as much aluminium or iron, otherwise chromium is incompletely precipitated. Adsorption of zinc by chromic hydroxide is even greater than by chromium phosphate, being (by Blum's method) 57 per cent. as against 26 per cent. by this method. Zinc is usually best separated by a direct method.⁴

(4) *Zinc Group Sulphides.*—No difficulty should be experienced with this group, provided that a fairly rapid stream of hydrogen sulphide is used, so as to get the solution saturated. Two minutes' passage of the gas (70° to 80° C.) at the rate of 500 ml. per minute with the use of the usual glass tube, will completely precipitate 1 mg. of cobalt, whereas a very slow stream may fail even after 30 minutes. Precipitation occurs in the order: ZnS, CoS, NiS; hence when zinc is present together with nickel or cobalt, the white zinc sulphide can be seen before the black sulphides begin to be precipitated.

If the passage of gas begins at 90° to 100° C., and the beaker is allowed to stand on a paper mat, the solution generally remains at 70° to 80° C. long enough

for the precipitation of the sulphides to be completed. Temperature is not very important when only zinc sulphide is to be precipitated; at 70° to 80° C. not more than 0.5 mg. of nickel or cobalt remains dissolved at pH 4.6 to 4.8; if the solution saturated with hydrogen sulphide is boiled for a few seconds before filtering, not more than 0.02 mg. of nickel or cobalt remains in solution.

The pH of the solution during precipitation of the zinc group is 4.6 to 4.8, and the solution should give a blue colour with bromocresol green. Phosphates of zinc, cobalt, nickel and manganese may separate from the hot solution at this pH, but can be disregarded; the first three are decomposed by the hydrogen sulphide treatment, and manganese phosphate is readily dissolved by the *N*/10 acetic acid wash solution. When much manganese is present it is advisable in quantitative analysis to wash until a few drops of washings give no reaction for manganese with persulphate. Under these conditions the sulphides retain only a negligible amount of manganese.

(5) *Manganese*.—The precipitate obtained is so characteristic as to make confirmation redundant; the red colour obtained by dissolving the manganese dioxide in potassium acid oxalate solution is a specific, sensitive and easily applied test. The separation of manganese alone is very useful; different amounts from 0.2 g. of manganese to fractions of 1 mg. have been successfully separated from unknown substances and confirmed by direct methods of determining manganese.

The hypochlorite should not be added too quickly, or traces of manganese may be oxidised to a soluble red compound which is only decomposed by reducing. Should a red filtrate be obtained, a few drops of nitrite should be added and the liquid boiled, when the red colour will disappear; for rough qualitative analysis the red colour can be disregarded. For quantitative work the filtrate should always be treated a second time with hypochlorite to ensure completeness of precipitation. During precipitation of the manganese dioxide there is usually a sudden darkening almost to black, and if the hypochlorite has been added slowly (one drop per second) and the addition then stopped, there is no red colour and precipitation is generally complete.

The *N*/10 nitric acid wash solution serves to dissolve any calcium phosphate that may be precipitated by the prolonged boiling; after being washed in this way the manganese dioxide contains at most traces of calcium or magnesium.

The manganese dioxide may be dissolved in hydrochloric acid or oxalic acid and determined as phosphate, or small amounts may be dissolved in dilute sulphuric acid with the aid of sodium nitrite or sulphite and estimated colorimetrically.

Other reagents, such as bromine and persulphates, have been tried in place of hypochlorite, but are not so satisfactory, precipitation being slower and often incomplete with larger amounts of manganese. This method of precipitating manganese avoids the use of large amounts of reducing substances, such as formates, and ammonium salts, as reagents.

(6) *Calcium*.—The amount of magnesium adsorbed by the calcium does not usually warrant re-precipitation of the calcium oxalate. The acid solution containing magnesium and oxalate, unlike an ammoniacal solution, has no tendency to deposit magnesium oxalate.

If calcium is to be determined, the precipitate is best collected in a Gooch

crucible, dried and ignited (not above 600° C.), carbonated by moistening with saturated ammonium carbonate, dried and re-ignited for 3 minutes at 500° to 600° C., and weighed as calcium carbonate. Results are very satisfactory, no sodium salts being retained by the precipitate.

(7) *Magnesium*.—The precipitate may be filtered off, preferably in a Gooch crucible, and, after thorough washing with 2 per cent. ammonia, treated with a few drops of strong ammonium nitrate solution, dried, ignited and weighed as $Mg_2P_2O_7$. Results are very satisfactory.

TABLE I
ADSORPTION OF METALS BY THE IRON-GROUP PHOSPHATES

| Taken g. | | Adsorption Per Cent. (on added metal) | Taken g. | | Adsorption Per Cent. (on added metal) |
|-------------|---------------------------|--|-------------|---------------------------|--|
| Cr | ≡ 0.250 of $CrPO_4$ | 26 | Cr | ≡ 0.250 of $CrPO_4$ | 5.5 |
| Zn | ≡ 0.250 of $Zn_3(PO_4)_2$ | | Co | ≡ 0.250 of $Co_3(PO_4)_2$ | |
| | 0.355 of P_2O_5 * | | | 0.355 of P_2O_5 | |
| Cr | ≡ 0.250 of $CrPO_4$ | 52 | Cr | ≡ 0.250 of $CrPO_4$ | 3.5 |
| Zn | ≡ 0.025 of $Zn_3(PO_4)_2$ | | Ni | ≡ 0.250 of $Ni_3(PO_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| Cr | ≡ 0.250 of $CrPO_4$ | 55 | Cr | ≡ 0.250 of $CrPO_4$ | 5.0 |
| Zn | ≡ 0.005 of $Zn_3(PO_4)_2$ | | Ca | ≡ 0.250 of $Ca_3(PO_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| Cr | ≡ 0.250 of $CrPO_4$ | 50-60 | Cr | ≡ 0.500 of $CrPO_4$ | 12 |
| Zn | ≡ 0.001 of $Zn_3(PO_4)_2$ | | Ca | ≡ 0.025 of $Ca_3(PO_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| Cr | ≡ 0.100 of $CrPO_4$ | 13 | Cr | ≡ 0.250 of $CrPO_4$ | 1.5 |
| Zn | ≡ 0.250 of $Zn_3(PO_4)_2$ | | Mg | ≡ 0.250 of $Mg_3(PO_4)_2$ | |
| | 0.200 of P_2O_5 | | | 0.355 of P_2O_5 | |
| Cr | ≡ 0.050 of $CrPO_4$ | 7.5 | Cr | ≡ 0.10 of $CrPO_4$ | 14 |
| Zn | ≡ 0.250 of $Zn_3(PO_4)_2$ | | Fe | ≡ 0.10 of $FePO_4$ | |
| | 0.142 of P_2O_5 | | Al | ≡ 0.10 of $AlPO_4$ | |
| | | | Zn | ≡ 0.16 of $Zn_3(PO_4)_2$ | |
| Cr | ≡ 0.250 of $CrPO_4$ | 8 | | 0.284 of P_2O_5 | 3.4 |
| Mn | ≡ 0.250 of $Mn_3(PO_4)_2$ | | Cr | ≡ 0.125 of $CrPO_4$ | |
| | 0.355 of P_2O_5 | | Al | ≡ 0.125 of $AlPO_4$ | |
| | | | Mn | ≡ 0.250 of $Mn_3(PO_4)_2$ | |
| Cr | ≡ 0.500 of $CrPO_4$ | 13 | | 0.355 of P_2O_5 | |
| Mn | ≡ 0.025 of $Mn_3(PO_4)_2$ | | Cr | ≡ 0.125 of $CrPO_4$ | |
| | 0.355 of P_2O_5 | | Fe | ≡ 0.125 of $FePO_4$ | |
| | | | Mn | ≡ 0.250 of $Mn_3(PO_4)_2$ | |
| Cr | ≡ 0.050 of $CrPO_4$ | 1.7 | | 0.355 of P_2O_5 | |
| Mn | ≡ 0.250 of $Mn_3(PO_4)_2$ | | | | |
| | 0.142 of P_2O_5 | | | | |
| | | | | | |

* In this and the subsequent tables P_2O_5 represents the total amount of phosphate taken in each of the tests. It does not correspond to the amount of chromium and zinc phosphates together, but is merely a suitable quantity in excess of the amount of the metal or metals of the iron group used in the experiment.

Tables I, II and III show percentage adsorptions (calculated on the added metal) by the iron-group phosphates. From the work of Lundell and Knowles⁵ it is evident that under the best conditions the ammonia method, phosphate being absent, gives results rather better for manganese, about the same for nickel and far worse for cobalt than this method. Under the same conditions (Blum's method, avoiding excess of ammonia) adsorption of zinc by iron is greater than by this new method; if excess of ammonia is added, adsorption of zinc is far less, but then the adsorption of nickel, cobalt and manganese becomes greater.

TABLE II

ADSORPTION OF METALS BY THE IRON-GROUP PHOSPHATES

| Taken g. | | Adsorption Per Cent. (on added metal) | Taken g. | | Adsorption Per Cent. (on added metal) |
|-------------|---------------------------------------|--|-------------|---------------------------------------|--|
| Al | 0.250 of AlPO_4 | 5 | Al | 0.050 of AlPO_4 | 1.5 |
| Zn | 0.250 of $\text{Zn}_3(\text{PO}_4)_2$ | | Zn | 0.250 of $\text{Zn}_3(\text{PO}_4)_2$ | |
| | 0.178 of P_2O_5 | | | 0.142 of P_2O_5 | |
| | | (at pH 3.2) | | | |
| Al | 0.250 of AlPO_4 | 7 | Al | 0.250 of AlPO_4 | 1.2 |
| Zn | 0.250 of $\text{Zn}_3(\text{PO}_4)_2$ | | Mn | 0.250 of $\text{Mn}_3(\text{PO}_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| | | (at pH 3.4) | | | |
| | | 9 | | | |
| Al | 0.250 of AlPO_4 | 11 | Al | 0.500 of AlPO_4 | 4 |
| Zn | 0.250 of $\text{Zn}_3(\text{PO}_4)_2$ | | Mn | 0.025 of $\text{Mn}_3(\text{PO}_4)_2$ | |
| | 0.710 of P_2O_5 | | | 0.355 of P_2O_5 | |
| | | (at pH 3.2) | | | |
| Al | 0.250 of AlPO_4 | 11 | Al | 0.250 of AlPO_4 | 1 |
| Zn | 0.100 of $\text{Zn}_3(\text{PO}_4)_2$ | | Co | 0.250 of $\text{Co}_3(\text{PO}_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| | | (at pH 3.2) | | | |
| Al | 0.500 of AlPO_4 | 25 | Al | 0.250 of AlPO_4 | 0.5 |
| Zn | 0.025 of $\text{Zn}_3(\text{PO}_4)_2$ | | Ni | 0.250 of $\text{Ni}_3(\text{PO}_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| | | | | | |
| Al | 0.500 of AlPO_4 | 20-30 | Al | 0.250 of AlPO_4 | <0.2 |
| Zn | 0.001 of $\text{Zn}_3(\text{PO}_4)_2$ | | Ca | 0.250 of $\text{Ca}_3(\text{PO}_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| | | | | | |
| Al | 0.100 of AlPO_4 | 3 | Al | 0.250 of AlPO_4 | <0.2 |
| Zn | 0.250 of $\text{Zn}_3(\text{PO}_4)_2$ | | Mg | 0.250 of $\text{Mg}_3(\text{PO}_4)_2$ | |
| | 0.200 of P_2O_5 | | | 0.355 of P_2O_5 | |

Table IV shows that this method compares well with the basic-acetate method as regards adsorption. The great increase in adsorption of zinc with increase in pH by both the basic-acetate and the new method should be noted. The results shown for the basic-acetate method are probably better than those normally obtained, because usually the pH attained is higher—about 5.3; also more phosphate than will combine with the iron-group metals is often present, causing phosphates of subsequent groups to separate with the iron-group phosphates. The addition of ferric chloride after these have been precipitated does very little

good; sufficient iron-group metals to combine with all the P_2O_5 must be present at the time of buffering; alternatively, the solution should be buffered to pH 3, not pH 5, ferric chloride then added to remove the excess of P_2O_5 and the solution then buffered to pH 5 to precipitate the excess iron as basic acetate. Smith¹ has shown how unreliable the usual basic-acetate method is for mixtures containing much phosphate.

TABLE III

ADSORPTION OF METALS BY THE IRON-GROUP PHOSPHATES*

| Taken g. | | Adsorption Per Cent. (on added metal) | Taken g. | | Adsorption Per Cent. (on added metal) |
|-------------|---------------------------|--|-------------|---------------------------|--|
| Fe | ≡ 0.250 of $FePO_4$ | 10 | Fe | ≡ 0.125 of $FePO_4$ | 10 |
| Zn | ≡ 0.250 of $Zn_3(PO_4)_2$ | | Al | ≡ 0.125 of $AlPO_4$ | |
| | 0.355 of P_2O_5 | | Zn | ≡ 0.250 of $Zn_3(PO_4)_2$ | |
| | | | | 0.355 of P_2O_5 | |
| Fe | ≡ 0.250 of $FePO_4$ | 3.0 | Fe | ≡ 0.500 of $FePO_4$ | 30 |
| Mn | ≡ 0.250 of $Mn_3(PO_4)_2$ | | Zn | ≡ 0.025 of $Zn_3(PO_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| Fe | ≡ 0.500 of $FePO_4$ | 6 | Fe | ≡ 0.250 of $FePO_4$ | 30 |
| Mn | ≡ 0.025 of $Mn_3(PO_4)_2$ | | Ba | ≡ 0.250 of $Ba_3(PO_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| Fe | ≡ 0.250 of $FePO_4$ | 0.7 | Al | ≡ 0.250 of $AlPO_4$ | 25 |
| Co | ≡ 0.250 of $Co_3(PO_4)_2$ | | Ba | ≡ 0.250 of $Ba_3(PO_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| Fe | ≡ 0.250 of $FePO_4$ | 0.3 | Cr | ≡ 0.250 of $CrPO_4$ | 52 |
| Ni | ≡ 0.250 of $Ni_3(PO_4)_2$ | | Ba | ≡ 0.250 of $Ba_3(PO_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| Fe | ≡ 0.250 of $FePO_4$ | <0.2 | Cr | ≡ 0.500 of $CrPO_4$ | 84 |
| Ca | ≡ 0.250 of $Ca_3(PO_4)_2$ | | Ba | ≡ 0.025 of $Ba_3(PO_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |
| Fe | ≡ 0.250 of $FePO_4$ | <0.2 | Cr | ≡ 0.250 of $CrPO_4$ | 12 |
| Mg | ≡ 0.250 of $Mg_3(PO_4)_2$ | | Sr | ≡ 0.250 of $Sr_3(PO_4)_2$ | |
| | 0.355 of P_2O_5 | | | 0.355 of P_2O_5 | |

* The last five sets of results show what would happen if barium and strontium were not first removed but were precipitated with the calcium.

Table V demonstrates the adsorption effects throughout the various groups in the new phosphate separation method.

I gratefully acknowledge my indebtedness to Mr. H. C. S. de Whalley for testing out this method at the Tate and Lyle Research Laboratory; also to Dr. Harold Toms of East Ham Technical College, where much of the practical work was done.

TABLE IV

ADSORPTION OF BIVALENT METALS BY THE BASIC-ACETATE METHOD
WITH PHOSPHATE PRESENT*

| Taken g. | pH by bromo-cresol green | Adsorbed by the basic-acetate precipitate | Adsorbed by new method Per Cent. (on added metal) |
|--|--------------------------------|--|--|
| Fe \equiv 0.250 of FePO_4 Zn \equiv 0.250 of $\text{Zn}_3(\text{PO}_4)_2$ $\text{P}_2\text{O}_5 \equiv$ 0.125 of FePO_4 } | 4.8 | 13.5 per cent. of the zinc | 10 |
| Fe \equiv 0.250 of FePO_4 Mn \equiv 0.250 of $\text{Mn}_3(\text{PO}_4)_2$ $\text{P}_2\text{O}_5 \equiv$ 0.125 of FePO_4 } | 4.8 | 4.5 per cent. of the manganese | 3.0 |
| Fe \equiv 0.250 of FePO_4 Ni \equiv 0.250 of $\text{Ni}_3(\text{PO}_4)_2$ $\text{P}_2\text{O}_5 \equiv$ 0.125 of FePO_4 } | 5.0 | 6 per cent. of the nickel | 0.3 |
| Fe \equiv 0.250 of FePO_4 Ba \equiv 0.250 of $\text{Ba}_3(\text{PO}_4)_2$ $\text{P}_2\text{O}_5 \equiv$ 0.125 of FePO_4 } | 5.0 | 14 per cent. of the barium | none |
| Al \equiv 0.250 of AlPO_4 Zn \equiv 0.250 of $\text{Zn}_3(\text{PO}_4)_2$ $\text{P}_2\text{O}_5 \equiv$ 0.125 of AlPO_4 } | 4.4 | 6 per cent. of the zinc | 8 |
| Al \equiv 0.250 of AlPO_4 Zn \equiv 0.250 of $\text{Zn}_3(\text{PO}_4)_2$ $\text{P}_2\text{O}_5 \equiv$ 0.125 of AlPO_4 } | 4.8 | 15 per cent. of the zinc | 8 |
| Al \equiv 0.250 of AlPO_4 Mn \equiv 0.250 of $\text{Mn}_3(\text{PO}_4)_2$ $\text{P}_2\text{O}_5 \equiv$ 0.125 of AlPO_4 } | 4.4 | 0.5 per cent. of the manganese | 1.2 |
| Al \equiv 0.250 of AlPO_4 Ni \equiv 0.250 of $\text{Ni}_3(\text{PO}_4)_2$ $\text{P}_2\text{O}_5 \equiv$ 0.125 of AlPO_4 } | 4.6 | 1.3 per cent. of the nickel | 0.5 |
| Fe \equiv 0.10 of FePO_4 Al \equiv 0.10 of AlPO_4 Cr \equiv 0.10 of CrPO_4 $\text{P}_2\text{O}_5 \equiv \frac{1}{2}$ of the Fe, Al and Cr | 4.3 | 20 per cent. of the zinc | 14 |
| Fe \equiv 0.250 of FePO_4 Co \equiv 0.250 of $\text{Co}_3(\text{PO}_4)_2$ $\text{P}_2\text{O}_5 \equiv$ 0.125 of FePO_4 } | 4.7 | 4.0 per cent. of the cobalt | 0.7 |

* The method employed for these basic-acetate separations was that of Treadwell and Hall, Vol. II, 1930; 1.0 to 1.2 g. of crystalline sodium acetate was used.

TABLE V

ADSORPTION EFFECTS THROUGHOUT THE GROUPS IN THE NEW
PHOSPHATE SEPARATION METHOD

| Taken g. | | Adsorption, etc. |
|---|---|--|
| Zn \equiv 0.200 of ZnS | } | 1.4 per cent. of the manganese carried down by the zinc sulphide. |
| Mn \equiv 0.200 of MnS | | |
| 0.142 of P_2O_5 | | |
| Ni \equiv 0.200 of NiS | } | 0.5 per cent. of the manganese carried down by the nickel sulphide. Not more than 0.25 mg. of nickel remained in the filtrate from the nickel sulphide. |
| Mn \equiv 0.200 of MnS | | |
| 0.142 of P_2O_5 | | |
| Co \equiv 0.200 of CoS | } | 0.3 per cent. of the manganese carried down by the cobalt sulphide. Not more than 0.25 mg. cobalt remained in the filtrate from the cobalt sulphide. |
| Mn \equiv 0.200 of MnS | | |
| 0.142 of P_2O_5 | | |
| Mn \equiv 0.200 of MnO | } | Not more than 0.5 per cent. of the calcium retained by the manganese dioxide. Obtained Ca \equiv 0.1986 g. of CaO. " Mn \equiv 0.2017 g. of MnO. |
| Ca \equiv 0.200 of CaO | | |
| 0.142 of P_2O_5 | | |
| Zn \equiv 0.200 of ZnO | } | Not more than 0.2 per cent. of the calcium carried down by the zinc sulphide. |
| Ca \equiv 0.200 of CaO | | |
| 0.142 of P_2O_5 | | |
| Ca \equiv 0.150 of CaO | } | 1.5 per cent. of the magnesium carried down by the calcium oxalate. Obtained 0.1530 g. of CaO. " 0.1480 g. of MgO. |
| Mg \equiv 0.150 of MgO | | |
| 0.142 of P_2O_5 | | |
| Ba \equiv 0.350 of $BaSO_4$ | } | 3 per cent. of the iron adsorbed by the barium sulphate. |
| Fe ⁺⁺⁺ \equiv 0.242 of Fe_2O_3 | | |
| 0.213 of P_2O_5 | | |
| Ba \equiv 0.250 of $BaSO_4$ | } | After reduction of the iron by hydriodic acid 0.2 per cent. of the iron adsorbed by the barium sulphate. Obtained 0.2517 g. of $BaSO_4$. |
| Fe ⁺⁺⁺ \equiv 0.250 of $FePO_4$ | | |
| 0.100 of P_2O_5 | | |
| Sr \equiv 0.151 of $SrSO_4$ | } | 1 per cent. of the iron adsorbed by the strontium sulphate. Filtrate contained not more than 1 mg. of strontium. |
| Fe ⁺⁺⁺ \equiv 0.242 of Fe_2O_3 | | |
| 0.213 of P_2O_5 | | |
| Sr \equiv 0.010 of SrO | } | After reduction with hydriodic acid. $SrSO_4 \equiv$ 0.0094 g. of SrO obtained containing 0.0002 g. of Fe_2O_3 . |
| Fe ⁺⁺⁺ \equiv 0.500 of $FePO_4$ | | |
| 0.142 of P_2O_5 | | |
| Sr \equiv 0.010 of SrO | } | $SrSO_4 \equiv$ 0.005 g. of SrO obtained. |
| Fe ⁺⁺⁺ \equiv 0.500 of $FePO_4$ | | |
| 0.142 of P_2O_5 | | |
| Sr \equiv 0.050 of $SrSO_4$ | } | 0.041 g. of $SrSO_4$ obtained. |
| Ca \equiv 0.500 of $CaSO_4$ | | |
| 0.100 of P_2O_5 | | |
| Sr \equiv 0.250 of $SrSO_4$ | } | 5 minutes' heating in bath gave 0.240 g. of $SrSO_4$. 15 " " " " " 0.245 g. " " 30 " " " " " 0.249 g. " " |
| 0.100 of P_2O_5 | | |
| | | |

REFERENCES

1. T. B. Smith, *J. Chem. Soc.*, 1933, 253.
2. M. E. Haring and M. Leatherman, *J. Amer. Chem. Soc.*, 1930, **52**, 5135.
3. M. E. Haring and B. B. Westfall, *id.*, 1930, **52**, 5141.
4. H. A. Fales and G. M. Ware, *id.*, 1919, **41**, 487; also their textbook, "*Inorganic Quantitative Analysis*," Bell, pp. 250-257 (1928).
5. G. E. F. Lundell and H. B. Knowles, *J. Amer. Chem. Soc.*, 1923, **45**, 676.
6. J. Mellor, "*Inorganic Chemistry*," Vol. XI, p. 374 (1931).

18, STRATTON DRIVE
BARKING, ESSEX

October, 1939

The Peroxide Accumulation Rate of Oils— Relationship to Mackey Test Results and Oxidation on Textiles

By W. GARNER, M.Sc., F.T.I., AND M. ELSWORTH, B.Sc., A.I.C.

THE work described below relates to esters (especially glycerides) of fatty acids, and some of the conclusions reached are not altogether applicable to fatty acids.

When oils (*e.g.* olive oil) are employed for fibre lubrication in wool-combing, 3 per cent. of oil is applied to the wool. The surface area of the fibres in 1 lb. of wool is approximately 70 square yards, and 1 lb. of oil is therefore spread over a maximum surface of about half-an-acre. The conditions are very favourable to rapid oxidation; this is necessary in order to produce fission products which are pale in colour and easily scoured out, but in certain circumstances it may result in the formation of resinous or paint-like polymers which are yellowish or brownish in colour and difficult to remove from the fibre by an alkaline soap scour. In some instances the oxidation (which is exothermic in character) may be so rapid as to cause spontaneous ignition of the oiled fibres. The suitability or otherwise of an oil for textile use cannot be predicted from the usual analytical data such as the iodine value, amount of unsaponifiable matter, etc. For textile and fire insurance purposes, it has been the custom to rely on the evidence furnished by the Mackey test.^{1,2}

The four main factors influencing the rate of oxidation of a non-drying oil are: (a) the surface/mass ratio of the oil during the experiment; (b) the activity of oxidising catalysts present; (c) the activity of antioxidants present; (d) the temperature conditions.

The chemical constitution of the oil is of very minor importance,^{3,4} compared with the catalyst-antioxidant balance. Temperature rise in the Mackey apparatus is probably due to heat liberated during peroxide formation and decomposition, both these reactions being accelerated by rise in temperature. For reasons to be discussed elsewhere,⁵ the rate of accumulation of peroxides (*i.e.* formation rate minus decomposition rate) in an oil which is exposed to light and air should be a measure of the effective activity (*i.e.* catalyst activity minus antioxidant activity) of the oxidising catalyst content.

The peroxide-content *per se* of an oil has been proposed as a criterion of the safety of a textile oil, but this appears to us to be of no significance. A given peroxide-content may be due to slow accumulation over a long period, the effective catalyst activity being low; or to rapid accumulation over a short period, the effective catalyst activity being high; it is assumed that other influencing conditions (light and heat) are equal. Conversely, two oils may show the same degree of effective catalyst activity, but have different peroxide-contents due, for example, to difference in age.

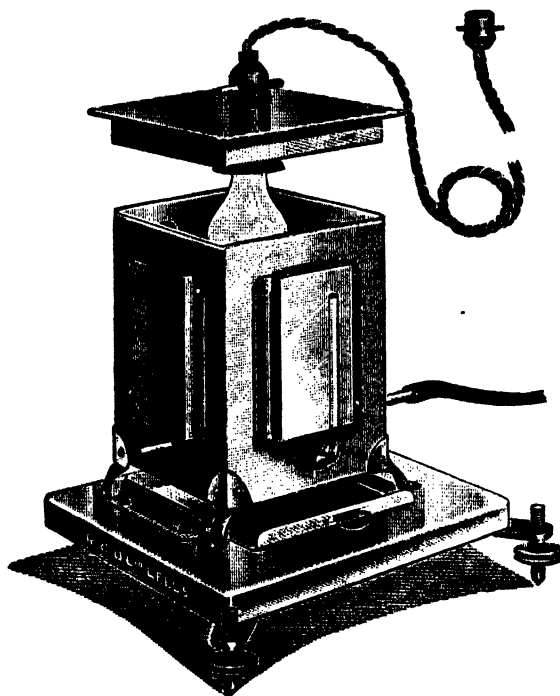


Fig. 1

The peroxide-content of an oil is a record of past history rather than an indication of future behaviour (see Table I), whilst the peroxide accumulation rate measures the liability of the oil sample to oxidise rapidly.

TABLE I

| Oil | Description | Peroxides | | Mackey test | | |
|---------|---|-----------|--------|--------------|-----------------------|--------------|
| | | P.I.V. | P.A.R. | °F at 5 hrs. | Mins. to reach 400°F. | Iodine value |
| Arachis | raw | 6.1 | 54.6 | — | 95 | 92.4 |
| " | Nilox—processed | 0.5 | 1.6 | 215 | — | 86.5 |
| Olive | Syrian .. 5 per cent. | 5.7 | 37.4 | — | 120 | 84.0 |
| " | Nilox | 0.6 | 1.2 | 210 | — | 84.3 |
| " | Malaga salad .. | 8.3 | 15.6 | 224 | — | 82.9 |
| " | African | 56.9 | 51.3 | — | 113 | 84.1 |
| " | " stored in dark 8 months to destroy peroxides .. | 0.5 | 35.7 | — | 110 | 85.1 |

The apparatus shown in Fig. 1 (p. 348) was therefore devised to measure the peroxide accumulation rate (P.A.R.). It consists of a tray carrying four shallow glass dishes (3-inch square Petri dishes). Oil is placed in one of the dishes, which is then exposed to the light from a 100-watt electric lamp, the heat rays being reduced by means of a 2-inch layer of running cold water. Under these conditions peroxides are usually formed quite rapidly; the difference between the amount of peroxide present before and after exposure is a measure of the peroxide accumulation rate.

The details of the test are as follows:—The peroxides in 1 g. of the oil sample are determined by the method of Lea,⁴ but calculated to give the number of mg. of iodine liberated per 1 g. of oil, this being termed the peroxide iodine value (P.I.V.). A peroxide iodine value of 1 indicates that roughly 1 mol. in 2000 is a peroxide (with ethyl oleate). Ten g. of the oil are then weighed into one of the Petri dishes and exposed to light in the apparatus for 48 hours. The P.I.V. is determined on the exposed sample, and the difference between exposed and unexposed value is termed the peroxide accumulation rate.

The variables in the test are:—(a) the four positions in the tray (See Table II); (b) the weight of oil per tray (*i.e.* surface/mass ratio) (See Table II); (c) the intensity of the light (See Table II); (d) the temperature of the oil (See Table II); (e) the method of determining peroxides; (f) time (See Table III); (g) oxidising catalyst activity (See Tables IV and V); (h) antioxidant activity (See Table V).

TABLE II

| Experiment No. | Variations from standard conditions | Initial P.I.V. | P.A.R. of oil in position | | | |
|----------------|---|----------------|---------------------------|------------------|------------------|------------------|
| | | | 1 | 2 | 3 | 4 |
| 1 | None | 1.2 | 16.6 | 17.4 | 19.2 | 17.6 |
| 2 | 60-Watt lamp instead of 100— Air current blown over dishes | 1.2 | { 8.2 — | { 7.9 — | 8.9 | 8.5 |
| 3 | Amount of oil in dish .. | 1.2 | 18.4 (5 ml.) | 17.2 (10 ml.) | 15.2 (20 ml.) | 11.8 (40 ml.) |
| 4 | Standing water; mean temp. 35° C. | 1.2 | 20.8 | 17.5 | 19.5 | 18.2 |
| 5 | No light | 1.2 | 1.3 | 1.3 | 1.2 | 1.3 |

TABLE III

| Hours | Iodine value | Fall in I.V. from previous reading | Peroxide iodine value | Rise in P.I.V. from previous reading |
|-------|--------------|------------------------------------|-----------------------|--------------------------------------|
| 0 | 83.3 | — | 27.2 | — |
| 8 | 82.3 | 1.0 | 33.0 | 5.8 |
| 16 | 80.3 | 2.0 | 38.6 | 5.6 |
| 32 | 80.3 | 0.0 | 42.0 | 3.4 |
| 40 | 78.9 | 1.4 | 48.0 | 6.0 |
| 48 | 78.4 | 0.5 | 53.2 | 5.2 |
| 60 | 74.6 | 3.8 | 69.2 | 16.0 |
| Total | | 8.7 | | 42.0 |

Note I. The 60-hour product heated *in vacuo* at 170° C. for 3 hours had I.V. 79.6, and P.I.V. 20, showing that peroxide formation is to some extent reversible.

Note II. A fall of one unit in iodine value corresponds with an increase of 4.8 in P.I.V.; this relationship indicates $O_2 = I$. The expected relationship is $O_2 = I_2$.

TABLE IV

| Oil | Ferric oleate added Per Cent. | Initial P.I.V. | P.A.R. |
|-----------------|-------------------------------|----------------|--------------|
| Ethyl oleate .. | 0.0 0.1 | 2.0 2.8 | 8.0 24.5 |
| Olive oil .. | 0.0 0.1 | 7.0 7.4 | 22.3 66.5 |

TABLE V

| Section | Oil | Treatment or addition | Peroxides | | Mackey test | | Storage of oiled wool | | | | | |
|---------|--------------------------|---|-------------------|--------|---------------------|-----------------------|-----------------------|--|------------------------------|-----------------|---|----------------------------------|
| | | | Initial P.I.V. | P.A.R. | °F. at 5 hrs. | Mins. to 400°F. | Iodine value | | | Free fatty acid | | |
| | | | | | | | Initial | After 7 days' normal storage, drop | Drop in ageing test | Initial | Increase after 7 days' normal storage | Increase in ageing test |
| A | Olive | None | 2.2 | 28.1 | — | — | 84.8 | 4.4 | 22.3 | 3.0 | 1.2 | 3.4 |
| | „ | 0.01% ferric oleate | 2.1 | 56.7 | — | — | 84.8 | — | 25.0 | 3.0 | 1.2 | 4.4 |
| | „ | 0.25 per cent. β - naphthol .. | 2.3 | 10.6 | — | — | 85.0 | 5.2 | 19.1 | 3.2 | 0.2 | 2.1 |
| | „ | Nilox-processed .. | 2.2 | 0.4 | — | — | 84.4 | 4.5 | 11.0 | 3.1 | 0.1 | 1.3 |
| B | Olive | None | 7.04 | 33.3 | — | 109 | 85.4 | — | 31.7 | 3.0 | — | 6.1 |
| | „ | Nilox-processed | 1.8 | 4.2 | 234 | — | 84.8 | — | 15.6 | 3.1 | — | 1.3 |
| | Arachis | None | 4.8 | 51.8 | — | 65 | 92.7 | — | 54.2 | 3.8 | — | 7.4 |
| | „ | Nilox-processed ^{7,8} | 0.2 | 3.2 | 210 | — | 87.3 | — | 11.8 | 0.1 | — | 2.2 |
| | Ester Oil S ⁹ | Nilox-processed .. | 8.9 | 4.1 | 222 | — | 42.7 | — | 10.0 | 3.1 | — | 2.6 |

Variables (a), (b) and (e) can be controlled exactly, and variations in (d) under normal laboratory conditions do not affect the results; variations in (c) are likely to be serious (owing to such causes as voltage drops in the mains supply, ageing of the bulb, deposits in the water-tray); hence a control test on a standard oil should always be included.

It should be pointed out that most antioxidants of the amine or phenol types are liable themselves to develop coloured oxidation products, and this may result in the production of stains on textile fibres. Moreover, once the effectiveness of an antioxidant is exhausted, the oil will become oxidised according to its nature; *e.g.* cotton seed or linseed oils may be stabilised by β -naphthol to give an excellent P.A.R. or Mackey test, but would yet be very unsuitable for textile purposes; this applies also to an olive oil containing considerable amounts of catalysts, but stabilised by additions of antioxidant. Antioxidants merely delay the onset of the oxidation, but do not affect the type of end-product; an oil yields the same type of oxidation product when antioxidant is present (though more slowly) as when it is absent.¹⁰

Table V indicates the parallel relationship between the P.A.R., the Mackey test, and the rate of oxidation when oiled wool sliver holding 3 per cent. of oil was (a) subjected to air at room temperature for 7 days, and (b) subjected to an "ageing test" (consisting of storing for seven days in a moist atmosphere at 90°C.). It is clear that there is qualitative agreement between the P.A.R., Mackey test, normal oxidation and ageing test.

SUMMARY.—(1) The oxidation of an oil should be considered from two aspects which are almost completely unrelated, *viz.*: (a) the *rate of oxidation*, which depends upon the catalyst/antioxidant balance; (b) the *type of end product*, which depends upon the amount of di- and poly-ethenoids present.

(2) The P.A.R. indicates clearly the existing catalyst—antioxidant balance: large P.A.R. figures go with bad Mackey tests, and rapid oxidation in use on textiles.

(3) The P.A.R. does not give any reliable indication of: (a) the amount of oxidising catalysts, as catalyst activity may be masked by antioxidants; (b) the type of end-product of oxidation (this may be examined by means of the alkaline ageing test¹⁰).

(4) A high P.A.R. always indicates an oil unsuitable for textile purposes, but a low P.A.R. does not necessarily indicate suitability, as very unsaturated oils can be prepared with low rates of oxidation.

(5) The P.A.R. seems to afford the same kind of information as the Mackey test and is easier to carry out consistently. Further, direct comparison can be made of two, three, or four oils under identical conditions. Both tests should be regarded as a means of condemning bad oils rather than of approving good ones. A good result may mean either: (a) the oil is free from oxidising catalysts, or (b) the catalyst activity has been swamped by antioxidants (an undesirable state of affairs).

(6) The P.I.V. of an oil bears no direct relationship to the iodine value, rate of oxidation in use, Mackey Test, P.A.R., or catalyst/antioxidant balance, and is almost useless as a means of evaluating textile oils.

REFERENCES

1. W. McD. Mackey, *J. Soc. Chem. Ind.*, 1896, **15**, 90.
2. W. Garner and W. Leach, *ANALYST*, 1936, **61**, 337.
3. W. Garner, *id.*, 1936, **61**, 519.
4. ——— *J. Text. Inst.*, 1937, **28**, 57–68.
5. ——— *J. Soc. Dyers Col.* (in press).
6. Lea, *Proc. Roy. Soc.*, 1931, 175.
7. *Wool Record*, 1939, April 6th.
8. Wool Industries Research Association Special Report on Nilox Arachis Oil.
9. Wool Industries Research Association Special Report on Ester Oil.
10. W. Garner, *ANALYST*, 1940, **65**, 219.

LISTER & CO., LTD.

BRADFORD

March, 1940.

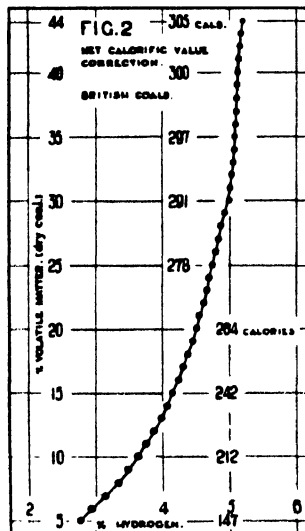
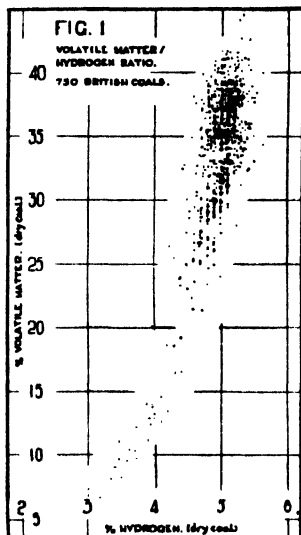
Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

THE NET CALORIFIC VALUE OF COALS

THE analyses normally required to control the quality of coal consignments are the proximate analysis, the sulphur-content and the gross calorific value. Analysts are frequently asked to provide, in addition, the net calorific value. This requires a further analytical determination, that of the hydrogen-content of the coal.

The examination of some seven hundred and fifty analyses of representative British coals indicates that it is possible to calculate the net calorific value by the use of the data available in the proximate analysis and without the additional determination of the hydrogen-content.



The relationships between carbon, hydrogen, volatile matter and gross calorific value have been noted and examined in detail by Seyler.¹ Recently the same investigator² has shown that the four macro-petrological constituents of coal—vitrain, clarain, durain and fusain—are aggregations of one or more micro-petrological units or "macerals," analogous to the minerals of inorganic rock types. Seyler² has also been able to demonstrate, by the examination of selected coals, that the micro-petrological units—vitrinite, fusinite, exinite and resinite, differ in chemical character; for example, macerals of different type but of the same elementary analysis vary in volatile matter content.

Industrial supplies of coal are composed mainly of clarain and durain—macro-constituents which are themselves mixtures of varying amounts of the macerals, vitrinite, fusinite, exinite and resinite. The hydrogen-content of coal samples of the same volatile matter content will therefore vary within limits according to the types and the amounts of the macerals present in each sample.

The variation in the ratio of volatile matter to hydrogen-content at any one value for volatile matter content may be judged from the graph in Fig. 1, in which the hydrogen-content on the dry basis is plotted against volatile matter for some

750 British coals. The examination of this large number of analyses has been made possible by the publication in the Technical and Survey Papers of the Fuel Research Board of full analyses of representative seam and commercial coal samples from most British coalfields other than South Wales. The data for low volatile South Wales coals have been abstracted from analyses made at University College, Cardiff.

The average hydrogen-content at any one specific volatile matter content is given by the graph of Fig. 2, drawn as a smoothed curve based on the computed average hydrogen values at 1 per cent. volatile matter intervals. In Fig. 2 are also shown the average net calorific value corrections at stated volatile matter intervals. Fig. 2 may also be used conveniently when directly determined hydrogen values are available.

A study of the data used for the construction of Figs. 1 and 2 shows that over 50 per cent. of the values for hydrogen-content are within ± 0.1 per cent. of the average value as read from Fig. 2, 78 per cent. within ± 0.2 per cent., 93 per cent. within 0.3 per cent., and 99 per cent. within ± 0.5 per cent. In terms of net calorific value corrections, 0.1 per cent., 0.3 per cent., and 0.5 per cent. hydrogen are respectively 5, 15 and 26 calories. If an experimental variation of the order of 25 calories is accepted as reasonable in the direct determination of the gross calorific value, then the graphical method of calculating the net calorific value by means of the volatile matter value is fully justified.

This method of calculation of the net calorific value may not be applicable to coals from overseas. In ninety analyses of representative American coals, published by Haslam,⁴ the hydrogen percentage derived from the volatile matter by the use of Fig. 2 is consistently lower than the determined value, the difference being as high as 2.0 per cent. with some high volatile American bituminous coals. It may be that the perhydrous macerals, exinite and resinite, occur in greater quantity and with greater variation in typical coals of the U.S.A.

T. EVANS

REFERENCES

1. C. A. Seyler, *Proc. South Wales Institute of Engineers*, April, 1900.
2. ——— *Fuel*, Jan., 1936.
3. ——— *Proc. South Wales Institute of Engineers*, Jan., 1938.
4. R. T. Haslam and R. P. Russell, "*Fuels and their Combustion*," New York, 1926, p. 68.

DEPARTMENT OF METALLURGY AND FUEL TECHNOLOGY
UNIVERSITY COLLEGE, CARDIFF
May, 1940

Notes from the Reports of Public Analysts

The Editor would be glad to receive the Annual or other Reports of Public Analysts containing matter of special interest to the Society. Notes made from such Reports are submitted to the Publication Committee.

MUNICIPALITY OF BANGALORE

ANNUAL REPORT OF THE PUBLIC ANALYST FOR THE YEAR 1939-40

THE Bangalore Prevention of Adulteration Act, 1937, came into operation in October, 1938, and the analytical work is carried out in the Municipal Laboratory, Civil and Military Station, Bangalore. During the last three months of 1938 the total number of samples examined was 787, comprising 423 of milk, 284 of ghee, 90 of butter and 10 of tea. Of these samples, 57 per cent. were adulterated, the percentage for each class being milk 47, ghee 73, and butter 60. All the samples of tea were genuine.

The milk sold in the Station is derived from the buffalo and the cow, but the latter is preferred, as in other parts of the country. Adulteration of cows' milk with buffalo milk is therefore very prevalent, in addition to the usual admixture with water. A small percentage of the adulterated samples contained separated milk, and in one sample boiled and fermented milk (commonly known as buttermilk in Bangalore) was detected.

The adulterated samples of ghee and butter contained various amounts of arachis oil, sesame oil, coconut oil and foreign animal fats. In some samples the vegetable fats constituted nearly 95 per cent. of the mixtures.

G. NARASIMHA MURTY

Department of Scientific and Industrial Research

METHODS FOR THE DETECTION OF TOXIC GASES IN INDUSTRY

ARSINE*

OCCURRENCE.—Arsine (arsenic trihydride) occurs in potentially dangerous concentrations in many industries, notably the manufacture of zinc chloride and sulphate and the smelting of arsenical ores; also in electroplating and galvanising works and in the manufacture of dyestuffs and of hydrochloric and sulphuric acids.

POISONOUS EFFECTS.—There is nearly always some delay—sometimes a day or two—in the onset of symptoms. At first these are usually indefinite; there may be severe headache, giddiness, nausea and vomiting. In more severe cases the vomiting may be more pronounced, and the urine dark or stained with blood. After a day or two there is severe anaemia, and the skin becomes jaundiced.

Exposure for 1 hour to an atmosphere containing 1 part of arsine in 20,000 is dangerous, and exposure for 12 hours to a concentration of 1 in 100,000 may be fatal. There are no recorded figures as to the limits of concentration that may be regarded as harmless for continuous daily exposure, but there is evidence that repeated exposures to very low concentrations may have cumulative effects resulting in severe poisoning.

METHODS OF DETECTION.—Although silver nitrate test-paper is about 20 times as sensitive as mercuric chloride paper, it has the drawbacks that the stains vary in colour from yellow to black according to the temperature of drying; that they darken very rapidly, so that accurate matching is difficult, and that the papers do not keep well.

In the standard test adopted, the test-papers are made by immersing the strips in 5 per cent. mercuric chloride solution, drying them, and cutting off and discarding the ends.

The atmosphere under examination is drawn by means of a hand-pump of specified dimensions through the test-paper, and the resulting stain (if any) is compared within 5 minutes with the standard stains issued with the leaflet. The concentration of arsine is then found by reference to the colour chart, which shows the intensities of stains corresponding with 10 to 50 strokes of the pump. In this way concentrations of arsine down to 1 in 250,000 (0.013 mg. per litre) can be detected. The sample of the atmosphere is passed through lead acetate paper to absorb any traces of hydrogen sulphide before coming in contact with the mercuric chloride paper.

* Leaflet No. 9. H.M. Stationery Office. Price 2s. 6d. net. Further copies of the standard stains. Price 2s.; by post 2s. 3d.

FOOD INVESTIGATION

THE FUNCTION OF NITRATE, NITRITE AND BACTERIA IN THE CURING OF BACON AND HAMS*

THE method of manufacturing bacon by tank curing in this country has been described in detail by Callow (*Biochem. J.*, 1929, 23, 648); it differs in many respects from the methods used in Canada and the United States, particularly in the temperature of curing. Also, in 1925, the use of sodium nitrite was authorised in the United States, provided that the amount in the finished product did not exceed 200 p.p.m. Sodium nitrite is also permissible in France, Germany, Holland, the Argentine, and other countries; in this country its use is illegal.†

The object of the present investigation was to determine the relative importance of the various factors contributing to the cured flavour of bacon, and also to ascertain if the use of sodium nitrite in the English method would give a satisfactory product (*cf.* Osman Jones, *ANALYST*, 1933, 58, 140). The colorimetric method of Lewis and Blake (*Allen's Organic Analysis*, 5th Ed., Vol. IX, pp. 420, 432), in which the reagents consist of α -naphthylamine and sulphanilic acid in hydrochloric acid, was used for the determination of nitrite.

ACTION OF HEAT ON NITRITE IN BACON.—It was found that with initial concentrations of 30 to 589 p.p.m. of sodium nitrite the time required to destroy about half of the nitrite increased with the concentration, *e.g.* from 13 to 120 minutes. Hence the usual times of cooking are unlikely to reduce the nitrite-content from a high to a low value.

NITRITE AND FIXATION OF COLOUR.—In absence of oxygen nitroso-haemoglobin is stable in solution. This *residual* nitrite in the tissue appears unnecessary for the fixation of colour, although it may possibly help to preserve the colour of cut surfaces exposed to the air.

FORMATION OF NITRITE FROM NITRATE AFTER CURING.—Judging by the concentration of potassium nitrate in the pickle, lean bacon may contain up to about 0.5 per cent. of nitrate. In one experiment with sliced bacon stored at 10° C. the nitrite-content increased from 22 p.p.m. to 260 p.p.m.—520 p.p.m. after 9 days.

NITRITE, SODIUM CHLORIDE AND WATER IN COMMERCIAL BACON.—Forty samples of bacon of the best quality (including Canadian, Danish, English, Irish and Swedish) contained from 8 to 204 p.p.m. of nitrite (as sodium nitrite) with a mean value of 57. The sodium chloride ranged from 3.0 to 7.6 per cent.; mean, 5.5 per cent., and the water from 53.9 to 68.2; mean, 64.1 per cent.

NITRATE, NITRITE AND FLAVOUR.—Tests applied to 23 of the above-mentioned 40 samples of bacon showed no correlation between flavour and nitrite-content; bacon containing as little as 10 p.p.m. had a satisfactory flavour. So far as can be ascertained, the distinctive flavour of bacon and ham is due to a reaction between the nitrite and constituents of the tissue either during curing or cooking.

BACTERIAL PRODUCTS AND FLAVOUR.—Contrary to a view that is held, the tests indicated that a good bacon flavour is produced by the action of sodium chloride and nitrite on flesh constituents, and that neither the presence of nitrate nor the action of bacteria is essential.

BACTERIAL FLORA OF TANK-PICKLES AND BACON.—Commercial pickle gave a very high count of bacteria, almost entirely micrococci when grown on ordinary media. Nearly all reduced nitrate—some even in 25 per cent. solutions of sodium chloride. The number of micro-organisms isolated was about 10^6 per ml., of which

* Special Report No. 49. By J. Brooks, Ph.D., R. B. Haines, D.Sc., Ph.D., T. Moran, D.Sc., Ph.D., and J. Pace, Ph.D. H.M. Stationery Office, York House, Kingsway, London, W.C.2. April, 1940. Price 9d. net.

† The Minister of Health made Regulations on October 20th, 1939, and April 30th, 1940, whereby the addition of sodium or potassium nitrite to bacon or ham is now permissible (see *ANALYST*, 1939, 64, 882; 1940, 65, 359).

90 per cent. were viable only on special media. Commercial samples of bacon gave counts of 10^6 or more viable organisms per sq.cm. of superficial tissue; they were mainly micrococci or yeasts or both. It seems unlikely that the development of bacterial flora has any material influence on the flavour.

ADVANTAGES OF THE USE OF NITRITE.—The direct use of nitrite in curing would enable the nitrite concentration in the pickle to be more rigorously controlled. At the same time, other constituents (sodium chloride, nitrate, flavouring ingredients, etc.) could be varied as desired, and the pH could be controlled. The bacon would keep better owing to the reduction in the surface bacteria flora, and the temperature of curing could be reduced, since it would no longer be necessary to maintain a balanced microflora in the tank-pickle.

Institute of Brewing

REPORT OF BREWERY EFFLUENTS COMMITTEE*

IN 1938 the Turton (Lancashire) Urban District Council put forward, under the Public Health (Drainage of Premises) Act, 1937, draft bye-laws, in which it was proposed: (1) to limit the amount of solids in suspension in trade effluents to 20 grains per gallon; (2) to prohibit the presence of yeast; (3) to prohibit the presence of sugar. The Brewer's Society drew the attention of the Institute to these proposals, and suggested that an investigation should be made to ascertain reasonable limits for solids in suspension, yeast and sugar.

As the result of this request a Brewery Effluents Committee was formed, consisting of Messrs. Bernard M. Brown (*Chairman*), Julian L. Baker, F. P. Clift, Harold Heron, H. B. Hutchinson, H. R. Lyell, Sir Gilbert Morgan, and W. H. Bird (*Secretary*).

This Committee considered the evidence immediately available as to the constitution of brewery effluents, and also obtained the expert opinion of Dr. E. Arden. In March, 1939, a Draft Report was issued asking for comments and pertinent information, and 40 replies were received. Only 4 gave recorded figures, and these fell within the ranges already considered.

CONSTITUTION OF BREWERY EFFLUENTS.—*Suspended Solids.*—Figures ranging from 5 to 1100 grains per gallon were shown by single samples. In one recently-constructed brewery averaged samples gave figures ranging from 6 to 67 grains per gallon, with a mean value of 32 grains per gallon.

Yeast.—In the above-mentioned brewery averaged 24-hour samples gave figures ranging from 265 to 8607 yeast cells per ml., with a mean value of 1774 cells; these figures are equivalent to 1.2, 37.7 and 7.8 grains of dry yeast per gallon. Single samples from another brewery gave figures ranging from *nil* to 260,000 yeast cells per ml., corresponding with *nil* to 823 grains of dry yeast per gallon.

Sugar.—Averaged samples from one brewery contained from 1.7 to 16.1 grains of reducing sugar (expressed as glucose) per gallon. At another brewery seven of eight single samples were free from sugar, and one contained sugar equivalent to 150 grains of glucose per gallon.

RECOMMENDATIONS.—(1) *General Considerations.*—It is pointed out that such bye-laws as may be proposed under the 1937 Act apply only to effluents from new premises, from premises to which an addition has been made, or from which on any one day a greater quantity of effluent is discharged into the public sewers than was discharged on any one day in the year ending March 3rd, 1937.

This consideration, however, is subject to the proviso that bye-laws which impose regulations regarding the temperature and neutralisation of trade effluents,

* *J. Institute Brewing*, 1939, 45, 551-555.

and the provision of inspection chambers or manholes and meters will apply to effluents, even though the consent of the Local Authority is not required under the Act to the discharge of such effluents.

Before bye-laws can be applied to brewery effluents, it will be necessary, in order to obtain samples that will conform to any reasonable standard, either to bring all outlets from the premises to one point commanding the public sewer, where a system of proportional sampling can be operated; or to have collecting tanks where the whole effluent can be averaged before its discharge into the sewer. The Committee lays stress upon the point that in many existing breweries it would be difficult to establish either of these systems.

(2) *Suspended Solids*.—Considering it undesirable to suggest a limit that could not in fact be achieved in practice, the Committee is of the unanimous opinion that more data are required before a figure can be put forward. Inasmuch as facilities exist under the Act for arrangements between local authorities and industrial concerns for the disposal of effluents, the Committee recommends that use should be made of these facilities, as far as is possible, to arrive at a mutually acceptable figure.

(3) *Yeast*.—The prohibition of yeast in an effluent would make it practically impossible to carry on the business of brewing. The model bye-laws of the Ministry of Health recognise the necessity of admitting some yeast, since they suggest the prohibition of "yeast in excess of"

The Committee recommends the acceptance of Dr. Ardern's suggestion that yeast should be permitted up to the limit of the suspended solids when such figure shall have been decided; this is in effect a recommendation that the yeast clause should be deleted.

(4) *Sugar*.—The Ministry's model bye-laws recognise that prohibition of sugar would be a hindrance to brewing, since they contain the words "sugar in excess of" Before a limit could be fixed it would be necessary to define sugar and to agree upon a method of determination. The Turton draft bye-laws and most other proposed bye-laws contain provisions for making a charge, beyond agreed limits, to the industrial undertaking, based on the figure for "oxygen absorbed" from acid permanganate in 4 hours at 80° F. Since excessive sugar content will result in high "oxygen absorbed" figures and will be paid for accordingly, it will be to the interest of the brewer to keep the sugar-content of the effluent as low as possible. The Committee, therefore, recommends non-acceptance of the whole sugar clause.

(5) *Temperature*.—In the Public Health Act, 1936, the discharge of effluents at a temperature higher than 110° F. was prohibited. This restriction is removed in the Public Health (Drainage of Premises) Act, 1937, unless re-imposed by a bye-law. In the opinion of the Committee this temperature limit (110° F.) should be generally accepted for any part of an effluent whether averaged or not. It is mentioned that this restriction is in force under the Public Health (London) Act, 1936.

(6) *Neutrality*.—The pH of beer is approximately 4, and still lower for waste beer. Since beer acidity is entirely of organic nature, no serious objection could be taken to its presence in sewers. The Committee therefore recommends a toleration range of pH 3.5 to 12 for breweries where the effluent is not averaged.

(7) *Local Agreements*.—In the opinion of the Committee, provided that certain principles are accepted on both sides, much more satisfactory results are likely to be obtained by local agreement than by attempts to conform to limits arbitrarily imposed.

Ministry of Food

THE SAUSAGES (MAXIMUM PRICES) ORDER, 1940*

THE Minister of Food has made this Order in exercise of the powers conferred upon him by Regulation 55 of the Defence (General) Regulations, 1939. It contains 7 Articles, which may be summarised as follows:

1. In this Order:—"Beef Sausages" means sausages which are ordinarily known and sold as beef sausages, and include sausages the meat in which is not beef alone.
"Pork Sausages" mean sausages which are ordinarily known and sold as pork sausages and include sausages the meat in which is not pork alone.
"Kosher Beef Sausages" mean beef sausages manufactured as respects the meat therein from beef obtained from cattle slaughtered in accordance with the Jewish practice of slaughter.
"Meat content" means as respects any sausage the percentage of the weight of meat contained in that sausage relative to the total weight of the contents of the sausage.†
In relation to sausages:—"Grade A" means sausages or sausage meat having a meat content of not less than 70 per cent. "Grade B" means sausages or sausage meat having a meat content of not less than 45 per cent. and less than 70 per cent.
"Grade C" means sausages or sausage meat having a meat content of not less than 30 per cent. and less than 45 per cent.
2. This forbids the sale of sausages or sausage meat at a price exceeding the maximum wholesale or retail prices applicable to the respective descriptions given in the Schedule to the Order.
3. Sausages or sausage meat with a meat content of less than 30 per cent. may not be manufactured or sold.
4. Retailers must indicate by means of a ticket or label the grade and meat content of the sausages or sausage meat offered or exposed for sale; it shall be sufficient compliance with the requirements of this Article if the ticket or label is prominently displayed on a slab or tray on which the sausages or sausage meat are exposed for sale.
5. (1) In any prosecution for the sale of sausages or sausage meat with a meat content other than that indicated by the ticket or label mentioned in Article 4, it shall be a defence for the defendant to prove.
 - (i) that he purchased the sausages or sausage meat with a written warranty to the effect that the meat content was that indicated on the label or ticket, and
 - (ii) that he had no reason to believe at the time of the commission of the alleged offence that such meat was other than that so warranted.(2) A warranty shall only be a defence to such proceedings if the defendant has within seven days of the service of the summons sent to the prosecutor a copy of the warranty with a notice stating that he intends to rely upon it, and specifying the name and address of the person from whom he received it, and has also sent a like notice of his intention to that person.
- (3) A servant of the defendant is entitled to rely upon the provisions of this Article in the same way as his employer would have been if he had been the defendant.
- (4) The person by whom the warranty is alleged to have been given shall be entitled to appear at the hearing and to give evidence.
- (5) For the purposes of this Article any statement of grade or meat content in an invoice or similar document relating to any sausages or sausage meat mentioned in that invoice or document shall be deemed to be a written warranty that the sausages or sausage meat to which the statement relates are of the grade of meat content mentioned in the statement.
- (6) Infringements of this Order are offences against Regulation 55 of the Defence (General) Regulations, 1939.
- (7) This Order, which came into force on March 26th, 1940, may be cited as the Sausages (Maximum Prices) Order, 1940.

Dated March 18th, 1940.

H. L. FRENCH,
Secretary to the Ministry of Food.

* Statutory Rules and Orders, 1940. No. 394. Emergency Powers (Defence). Food. H.M. Stationery Office. Price 1d. net.

† For the determination of Meat Content see ANALYST, 1940, 65, 257.

Ministry of Health

THE PUBLIC HEALTH (PRESERVATIVES, ETC., IN FOOD) AMENDMENT REGULATIONS, 1940*

THE Minister of Health has made the following Regulations under the Food and Drugs Act, 1938:

- 1.—These Regulations and the Public Health (Preservatives, &c., in Food) Regulations, 1925 to 1927, shall be construed together and may be cited together as the Public Health (Preservatives, &c., in Food) Regulations, 1925 to 1940. They shall come into force on the date hereof.
- 2.—The Public Health (Preservatives, &c., in Food) Regulations, 1925 (a), as amended (b), shall be further amended as follows:
 - (1) The following additional proviso shall be inserted at the end of Article 4 (1) and at the end of Article 2 (1):

“(iv) The provisions of this Act shall not apply
(a) So as to prohibit the presence in bacon, ham and cooked meat of any added sodium or potassium nitrite.
(b) So as to prohibit the presence in any article of food of sodium or potassium nitrite introduced in the preparation of such article by the use of any bacon, ham or cooked pickled meat containing sodium or potassium nitrite.”
 - (2) The following paragraph shall be inserted after Article 4 (3):

“(4) No person shall manufacture for sale or sell any cooked pickled meat intended for human consumption, other than bacon or ham, which contains sodium or potassium nitrite in proportions exceeding two hundred parts per million calculated as sodium nitrite.”
 - (3) The following paragraph shall be inserted after Article 11 (2):

“No person shall import into England or Wales any cooked pickled meat intended for sale for human consumption, other than bacon or ham, which contains sodium or potassium nitrite in proportions exceeding two hundred parts per million calculated as sodium nitrite.”

(L.S.)

J. N. BECKETT,

Assistant Secretary, Ministry of Health.

British Standards Institution

THE following British Standard has been issued†:

NO. 895—1940. METHODS FOR THE MICROBIOLOGICAL EXAMINATION OF BUTTER.

In 1930 the Empire Marketing Board appointed a Committee to consider the standardisation of Methods for the Chemical and Bacteriological Analyses of Milk, Cheese and Butter Samples. This work was taken over by the British Standards Institution in 1933, and the continuance of the work of the Committee was confirmed on November 1st, 1934, at a meeting of the Dairy Standards Technical Committee, with the addition of two new members representing the Society of Public Analysts and Other Analytical Chemists. Methods for the bacteriological examination of butter were reviewed, and it was decided that to be of practical value the methods should be as few and as simple as possible, and should be reproducible, should reflect the commercial properties of butter, reveal faults in manufacture, and afford some measure of the keeping quality. Pathogenic organisms were regarded as outside their purview. A survey of the literature on the

* Statutory Rules and Orders, No. 633. Regulations dated April 30th, 1940. These Regulations supersede the provisional Regulations dated October 20th, 1939. H.M. Stationery Office, 1940. Price 1d. net.

(a) S.R. & O. 1925 (No. 775), p. 1375.

(b) S.R. & O. 1926 (No. 1557), p. 1177; 1927 (No. 577), p. 455.

† Obtainable from the Publications Department, British Standards Institution, 28, Victoria Street, London, S.W.1. Price 2s. net. Post free 2s. 2d.

bacteriological examination of butter showed that apparently no attempt had been made to establish the reproducibility of results when different workers were applying prescribed methods to the same sample of butter. The Committee decided to repair this deficiency.

The methods recommended are given under the following headings:—(A) GENERAL.—*Method of Sampling—Treatment of Sample—Apparatus—Diluent (Ringer's Solution)—Plating and Counting.* (B) MEDIA FOR THE DIFFERENTIAL ENUMERATION OF VARIOUS MICRO-ORGANISMS IN BUTTER.—*Yeasts and Moulds—Lipolytic (Tributyryl-splitting) Organisms—Caseolytic Organisms—Saccharolytic Organisms—Coliform Organisms.*

These methods are intended as the basis for observations comparing the counts of organisms of the various types with the commercial properties of the butter and revealing faults in manufacture, transport or storage. Those who use the methods are invited to communicate their results to the British Standards Institution.

The Committee recommends storage at 60° F. (as used by the National Mark Butter Committee) for the purpose of observation of keeping quality after cold storage.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Aromatic Principle in Bread. II. Isolation of the Aromatic Principle in Rye Bread. E. Komm and G. Lehmann. (*Z. Unters. Lebensm.*, 1940, **79**, 242–246.)—The bread used in the investigation was a highly flavoured regimental bread two or three days old. Steam-distillation was found unsatisfactory, owing to the high resistance of the bread-pulp, but methyl alcohol, ether, chloroform and acetone extracted the aromatic principle, and methyl alcohol free from acetone was chosen as the most convenient solvent. The finely-crumbled bread (40 kg. in 2000-g. portions) was extracted for 18 hours with 1500 ml. of boiling methyl alcohol. The brownish turbid extract (fraction F_1) was filtered, and a portion when allowed to evaporate at room temperature left a brownish viscous residue with a strong odour of bread. When the extract was distilled *in vacuo* it yielded a distillate (F_2) with an odour resembling that of whisky. This fraction was separated by distillation at ordinary pressure into 6 fractions (F_3 to F_8), at 12 mm. pressure into 4 fractions (F_9 to F_{12}), and at 4 mm. pressure into 10 fractions (F_{13} to F_{22}). None of these fractions indicated any concentration of the aromatic substances. By salting out and extracting F_2 with ether a fraction F_{23} was obtained, and this left on evaporation an acid residue (F_{24}) smelling strongly of bread. Attempts to isolate the aromatic body from this by extraction with solvents failed. F_{24} was neutralised, (F_{25}), saponified for 30 hours with sodium hydroxide, and finally neutralised with hydrochloric acid (F_{26}). This was fractionally distilled, and the fraction with the strongest odour (F_{28}) was further separated into 3 fractions (F_{29} to F_{31}) and F_{30} again into 3 fractions (F_{32} to F_{34}). All these fractions had a strong odour of bread, but the yield was small. F_{28} was evaporated, and the small residue was soaked up with filter-paper, which was then heated in a distillation flask. Three fractions were obtained, of which F_{37} had the strongest odour. The various fractions were examined for the presence of characteristic groups. Carboxylic acids were detected by the thionyl chloride test (Feigl, "*Qualitative Analyse mit Hilfe von Tüpfel Reaktionen*," Leipzig, 1939) in fractions 1 to 24 and 27. Oxy-acids were found in the same fractions by the ferric chloride test, and F_{28} gave a positive result in the Denigès test for lactic acid.

Acetic acid (Feigl's lanthanum nitrate test, *op. cit.*) was present in F_{24} . Phosphoric acid was found in fractions 1 to 3, 9, 13, 23, and 24. The first 30 fractions reduced Fehling's solution. Carbonyl groups were found in F_{28} by the test described in the next abstract. All the fractions answered to Feigl's test (*op. cit.*) for aldehydes with sodium bisulphite, and most of the fractions responded to Fischer and Penzoldt's test for aliphatic aldehydes with diazobenzenesulphonic acid (*Ber.*, 1884, 17, 572). Angeli's test for aromatic and aliphatic aldehydes (*Gazz. Chim. Ital.*, 1896, 26, 17) gave positive results with all fractions except F_{17} . Almost all fractions answered to Feigl's test (*op. cit.*) for $\alpha\beta$ -unsaturated and aromatic acids with sodium pentacyanoammine ferroate. None of the fractions responded to Legal's test for methyl ketones. Phenols could not be detected, but most of the fractions contained small amounts of higher alcohols. Fusel oil was found in fractions 5 to 8. Since the foregoing tests indicated the presence of unsaturated and aromatic aldehydes in many fractions, individual representatives of these classes were sought for. Zinke and Dietmann's test for furfuraldehyde with a mixture of alcoholic aniline and hydrochloric acid (Gattermann, "*Die Praxis Organischen Chemikers*," Berlin and Leipzig, 1933, p. 374) indicated a high concentration of that substance in F_{37} , the fraction with the strongest odour of the original bread. Most of the other fractions answered to this test. Fiehe's test for hydroxyfurfuraldehyde was given by fractions 8, 12, 27, 28 and 31. The investigation has been interrupted, but from the results already obtained it would appear that the aromatic principle most strongly concentrated in fraction 37 is a substituted furfuraldehyde derivative.

A. O. J.

Aromatic Principle in Bread. III. Colorimetric Estimation of Diacetyl, Acetoin and Butylene Glycol. E. Komm and J. Flügel. (*Z. Unters. Lebensm.*, 1940, 79, 246-250.)—Acetoin (acetylmethylcarbinol) forms a red colour on atmospheric oxidation in presence of 40 per cent. sodium hydroxide solution. Attempts to accelerate the oxidation by using more powerful oxidising agents failed to give a reaction suitable for quantitative purposes. Acetoin and butylene glycol are converted into diacetyl by suitable oxidising agents. Previous work on the estimation of creatinine by means of *m*-dinitrobenzoic acid (Komm and Pinder, *Z. Unters. Lebensm.*, 1939, 78, 113; Abst., ANALYST, 1940, 65, 229) suggested that a similar method might be applied to the colorimetric estimation of diacetyl. Experiments with a number of nitro-compounds showed that an alcoholic solution of *m*-dinitrobenzoic acid (1:3:5) was the most convenient reagent, yielding a violet to wine-red colour with cold solutions of diacetyl in presence of alkali. The course of the reaction is influenced by the temperature, the alkali concentration and the duration of reaction. A study of the effects of varying each of these factors in turn led to the following procedure:—The aqueous diacetyl solution (2 ml.) is mixed with 1 ml. of 5 per cent. sodium hydroxide solution, and 2 to 4 ml. of alcoholic dinitrobenzoic acid solution are added so as to form a separate layer. Standard solutions of diacetyl are treated in the same manner. All the reaction mixtures are shaken simultaneously, and the colours are compared after 20 minutes. The method gives satisfactory results with solutions of diacetyl of concentration greater than 1 part in 100,000. It was

used successfully for the estimation of diacetyl, acetoin and butylene glycol in leavened bread. Acetoin and diacetyl were estimated together after oxidation of the acetoin by means of ferric oxide in presence of sulphuric acid. Butylene glycol was converted into acetoin by oxidation with bromine, and then into diacetyl by oxidation with ferric chloride. For diacetyl and acetoin the procedure was as follows:—The finely crumbled bread (150 g.) was distilled in steam, and the distillate was oxidised as described, and again distilled, 60 ml. of distillate being collected. Two ml. of the distillate were used for the colorimetric estimation. For the estimation of butylene glycol, 150 g. of crumbled bread were covered with 300 ml. of water and 10 ml. of conc. sulphuric acid and distilled in steam until 5 litres had been collected. A 500-ml. portion of the distillate was warmed with 1 ml. of bromine on the water-bath for 30 minutes at 70° to 80° C. beneath a reflux condenser. The mixture was rapidly cooled, the bromine was removed by adding finely powdered ferrous sulphate and, after addition of 10 ml. of conc. sulphuric acid and 30 ml. of 30 per cent. ferric chloride solution, the liquid was distilled, 60 to 100 ml. of distillate being collected. The distillate (2 ml.) was used for the colorimetric estimation. Comparative determinations were made by the gravimetric nickel dimethylglyoxime method of Visser't Hooft and de Leeuw (*Biochem. Z.*, 1925, 161, 361). The colorimetric method gave somewhat higher results.

A. O. J.

Characterisation of Starch from Various Sources by means of Electrophoretic Fractionation. O. Dahl. (*Z. physiol. Chem.*, 1940, 263, 81–99.)—A 2 per cent. paste of the starch was treated for 2 to 3 days in an electrophoresis apparatus with an e.m.f. of 40 volts per cm. The sol produced was concentrated under reduced pressure to give a 1 per cent. solution (Amylose I). The gel (Amylopectin I) was diluted with water to re-form a paste, which was re-treated, thereby forming a second sol (Amylose II) and gel (Amylopectin II). A similar treatment of the second gel gave Amylose III and Amylopectin III. The organic phosphorus content of the six fractions prepared from each kind of starch was determined, and also the amount of phosphorylation produced by muscle phosphorylase in a given time was ascertained by measuring the usage of inorganic phosphate. It was found that the organic phosphorus contents of the starches of wheat, rice, maize and potato and of the amylopectin fractions prepared from them were 0.01 to 0.08 per cent., and that phosphorylation proceeded to approximately the same degree with all the starches and their amylopectin fractions. The yields of amylose were 16 to 18 per cent. for wheat-, maize- and potato-starch, and 9 per cent. for rice-starch. The amyloses did not contain any appreciable amount of organic phosphorus, and were phosphorylated to the extent of one-third to one-half the native starches and amylopectins, with the exception of potato-amylose, which was phosphorylated to the extent of only one-eighth. There appeared to be no relationship between the phosphorus-content and the degree of phosphorylation. The anomalous potato-amylose to some extent inhibited the phosphorylation of other substrates. The amylose and amylopectin fractions of wheat- and potato-starch were fractionally digested with α - and with β -amylase, and the extent to which each of the degradation products was phosphorylated was

ascertained. It was found that all the fractions obtained by α -amylase degradation quickly lost their ability to be phosphorylated, whilst the fractions treated with β -amylase did not.

F. A. R.

Reducing Power of Unripe Honey and Honey from Sugar-fed Bees.

R. F. Kardos. (*Z. Unters. Lebensm.*, 1940, 79, 258-262.)—The reducing power of honey towards chloramine, under the conditions laid down by Tillmans and Hollatz, was studied as a possible means of discriminating between different classes of honey. This reduction is slight and is due, not to the sugars, but to minute quantities of unknown substances. Ten ml. of 10 per cent. honey solution (10 g. in 100 ml. of solution) were oxidised with 20 ml. of $N/100$ chloramine solution after acidification with 3 ml. of 2 N acetic acid. After the reaction mixture had stood for 10 minutes, dilute sulphuric acid and potassium iodide were added, and the iodine liberated by the excess of chloramine was titrated with $N/100$ sodium thiosulphate solution. The chloramine consumption for sugar-fed honey was found to be 1.1 to 2.3 ml. of $N/100$ per g., which is lower than the range found for normal honey. Unripe honey resembled sugar-fed honey in many of its properties and had a similarly low reducing power (0.9 to 1.9 ml. of $N/100$ per g.). Two samples, although containing no added sugar-fed honey, had exceptionally high sucrose-contents (14.8 and 18.8 per cent.). From this it is concluded that not only sugar-fed honey, but also honey collected by bees brought up on sugar, may contain much sucrose. The reducing power of ripe normal honey ranged from 1.0 to 11.9 ml. of $N/100$ per g. Four samples of ripe, yellow spring honey showed very low reducing powers, and a series of spring honeys gave a range of 1.1 to 2.0. The reducing power should be used with caution as a proof of the presence or absence of sugar-fed honey in a given sample. The reducing power of summer and autumn honey is 3 to 5 ml. (or more) of $N/100$ per g. With sugar-feeding the reducing power is not more than 2 to 3 ml. of $N/100$ per g. In such instances sugar-feeding may be assumed from the lower figure, especially if the sucrose-content, diastatic power and protein-content are within normal ranges. In fresh spring honey a low reducing power does not necessarily indicate sugar-feeding. The first spring honey is separated very early in the year and is often unripe. In winter, when the bee does not collect honey, it apparently loses the power of depositing in the honey the small amounts of biologically important substances that are responsible for the reducing power. For this reason the earliest spring honey (*e.g.* acacia honey) resembles sugar-fed or unripe honey, whereas the "second" acacia honey collected and separated two or three weeks later approaches normal honey in composition. Spring honey that has been gathered by bees artificially fed with sugar during the winter resembles sugar-fed honey. It may be that the bees in inverting are "tired," and also that their brood does not receive correct nourishment. A further reason for the weakness of spring honey may be that the introduction of artificial comb into the hive stimulates comb-building and causes the production of larger amounts of correspondingly weaker honey. It is well-known that the sucrose-content of sugar-fed honey is in general higher than that of normal honey of the same kind, and that on standing the sucrose-content falls more slowly than that of normal honey.

A. O. J.

Estimation of Silver in Catadynised Vinegar and Fruit Cordials.

O. Noetzel. (*Z. Unters. Lebensm.*, 1939, **78**, 315–321.)—As the catadyn process of sterilising food is now widely used, especially for vinegar and fruit cordials, it is necessary in food control to ensure that the amount of silver present is not too high; it is considered that a quantity of 500 γ per litre is sufficient for sterilisation. For estimating the silver the method of Feigl (*Z. anal. Chem.*, 1928, **74**, 380; *Abst., ANALYST*, 1928, **53**, 615), which depends on the formation of a red silver compound with *p*-dimethyl-aminobenzylidene-rhodanine, is recommended. *Silver in vinegar.*—The organic matter is destroyed either by (1) ignition or (2) a wet process with sulphuric and nitric acids. (1) Evaporate 50 to 100 ml. of the sample in a porcelain crucible, ignite the residue and fuse it with 0.3 g. of a mixture of sodium carbonate and potassium nitrate (1 + 2.5). Dissolve the fused mass in 2 ml. of water and 3 ml. of nitric acid (sp.gr. 1.4) on the water-bath, and evaporate the solution to dryness. Dissolve the residue in 3 drops of nitric acid and 12 ml. of water, add ammonia in excess, boil and filter. Rinse the flask with dilute ammonia, neutralise the filtrate with dilute nitric acid, add 10 per cent. ammonia until the liquid is slightly alkaline, and make up to 30 or 50 ml. Add to an aliquot part 5 ml. of ether and a few drops of a 0.03 per cent. solution of *p*-dimethyl-aminobenzylidene-rhodanine in acetone. In presence of silver a red film of $\text{AgC}_{12}\text{H}_{11}\text{N}_2\text{OS}_2$ will be formed at the zone of contact of the liquids. The test is capable of detecting 2 γ of silver. A colorimetric estimation may be made by comparison with standards obtained with silver solutions prepared by diluting 4.65 ml. of *N*/10 silver nitrate solution plus 5 drops of nitric acid to 500 ml. (1 ml. = 100 γ of silver) or by diluting 1 part of this solution to 10 (1 ml. = 10 γ). (ii) For the wet process of destroying organic matter, evaporate 200 to 300 ml. of vinegar to 30 ml., add 30 to 40 ml. of nitric acid (sp.gr. 1.4), and boil until red fumes no longer appear. Add 6 to 7 ml. of conc. sulphuric acid, and boil with dropwise addition of nitric acid until the liquid is colourless. Cool, dilute with a 4-fold volume of water, add ammonia in excess (about 45 ml. of 10 per cent. ammonia solution in all), boil, filter, rinse the flask with dilute ammonia solution, neutralise (to litmus paper), add 1 drop of ammonia solution, and make up 100 ml. Estimate the silver colorimetrically as in (i).

Fruit cordials.—The wet process should be used; 60 to 70 ml. of nitric acid and 7 to 8 ml. of sulphuric acid are required. A shortened process depending on adsorption on activated charcoal may also be used. The fruit cordial (100 to 200 ml.) is diluted with an equal volume of water in a 500-ml. flask, shaken with 0.8 g. of activated charcoal and boiled for 45 minutes, so that its volume is reduced to about a half. After standing for 24 hours the mixture is filtered with the aid of suction in a Buchner funnel (6 cm. in diameter) containing a double layer of filter-paper. The filter and its contents are dried and ignited and the silver in the residue is estimated as described above.

D. A.

Mixed Unsaturated Glycerides of Liquid Seed Fats. Some "Non-Drying" Oils. **B. G. Gunde and T. P. Hilditch.** (*J. Soc. Chem. Ind.*, 1940, **59**, 47–53.)—The acetone solubility method of investigating the glyceride structure breaks down for liquid fats. In fatty non-drying oils, however, with oleic acid

as a preponderant component acid, elaidinisation (geometrical isomerisation) causes transformation of considerable proportions of mixed oleo-glycerides into corresponding mixed elaido-glycerides, which are solid at ordinary temperatures and resolvable to a considerable extent by crystallisation from acetone at 0° C. By using Bertrand's method with selenium at 220° C. as isomerising catalyst (*Chem. Weekblad*, 1936, 33, 3; cf. *ANALYST*, 1936, 61, 866) many objectionable by-products are eliminated, but the temperature causes destruction of linolic groups to an undesirable extent, so that figures deduced for the proportions of the component glycerides of the oils examined are of a definitely lower order of accuracy than those previously obtained for solid fats which can be directly fractionally crystallised. Application of the method to two olive oils (from Palestine and Italy), ground-nut and almond oils has indicated that when oleic acid forms less than about 60 per cent. of the total acids, as in ground-nut oil, while saturated and linolic acids are in the neighbourhood of 20 per cent. each, a considerable proportion of the oil may consist of monosaturated-mono-oleo-monolinoleo glycerides with small amounts of monosaturated-dioleins, linoleo-dioleins and triolein. When the oleic acid is much in excess of 60 per cent. of the total acids, as in olive and almond oils, and saturated and linolic acids each amount to less than 15 per cent., mono-oleo-glycerides are present, if at all, in very small proportions, and practically all the linolic acid is present as linoleo-dioleins. In such oils a fairly close approximation to the proportion of component glycerides can be obtained directly from the molar percentage proportions of the component acids by combining each minor component acid in the form of e.g. monopalmito-diolein or monolinoleo-diolein, the surplus of oleic acid being present as triolein, and allowance being made for any fully saturated glycerides present. In olive oils the amount of triolein probably rarely exceeds 35 per cent., and in some oils may be almost completely absent. The proportions of component glycerides, approximate, and regarded as probably present in almond oil are myristo-dioleins, 3, palmito-dioleins, 14, linoleo-dioleins, 52, and triolein 31 per cent. (mol.); in ground-nut oil, the combined proportions of the individual saturated acyl dioleins and acyl oleo-linoleins are given as palmito- 30, stearo- 11, arachido- 6, beheno- 7, and lignocero- 2 per cent. (mol.). The probable component glycerides of (a) Palestine olive oil and (b) Tuscany oil are given as tripalmitin (a) 2, (b) 2; myristo-diolein (a) 2, (b) 5; palmito-dioleins, (a) 26, (b) 38; stearo-dioleins, (a) 10, (b) 6; hexadeceno-diolein, (a) 3, (b) 5; linoleo-diolein (a) 26, (b) 38; triolein (a) 31; oleo-linoleo-palmitins, (b) 6 per cent. (mol.). The data for (b) are not wholly conclusive, and somewhat more mono-oleo-glycerides, together with a small proportion of triolein, might equally well be present. In both olive oils the proportion of triolein itself is far smaller than was formerly supposed.

D. G. H.

Determination of the Ascorbic Acid Content of Tablets. E. C. M. J. Hollman. (*Pharm. Weekblad*, 1930, 77, 393-400.)—The procedure recommended in the Supplement to the Dutch Pharmacopoeia for the determination of ascorbic acid is considered to be unsuitable for use with tablets, which usually contain talc, lactose and starch, and the following method is preferred:—The tablet is rubbed in 2 per cent. acetic acid solution until it is disintegrated, and the mixture

is diluted in a graduated flask to 500 ml. (presumably with the acetic acid.—J. G.). Five ml. of the solution are immediately pipetted into a porcelain dish, 15 ml. of a 3 per cent. trichloroacetic acid solution are added, and the mixture is back-titrated with a standard solution of 2,6-dichlorophenol-indophenol, with agitation, until a rose colour is produced and is permanent for 5 seconds. The indicator solution is prepared by dissolving 20 mg. of the 2,6-dichlorophenol-indophenol in 100 ml. of water, filtering the solution 24 hours later, and adding a little sodium bicarbonate to the filtrate. It should be renewed at least once a week, as if it is kept longer a turbidity develops and a dirty brownish-red colour is produced at the end-point. The importance of minimising the period which elapses between the preparation and titration of the solution of the sample was demonstrated in experiments with a mixture containing 100 mg. of pure ascorbic acid and 150 mg. each of lactose and of starch. Intervals of up to 4 hours were allowed to elapse before titration, and a progressive fall in the titration with lapse of time was found. Similar results were obtained whether water or 2 per cent. acetic acid was used, but when 8 commercial tablet preparations were compared from this point of view, the latter solvent always gave the higher results, and it is therefore recommended for use in the final method. The results indicate that formation into tablets does not cause any reduction in ascorbic acid content (Schou and Bennekou, *id.*, 1938, 75, 154; Stevens, *id.*, 1938, 75, 790). J. G.

Biochemical

Estimation of Small Amounts of Sulphur as Sulphates in Biological Fluids. A. D. Marenzi and R. F. Banfi. (*Biochem. J.*, 1939, 33, 1879–1889.)—An improved method of estimating sulphates in blood and urine has been devised, in which benzidine is used as precipitant and the benzidine is estimated with phosphotungstomolybdic acid. *Sulphur in blood.*—To 1 volume of whole blood, serum or plasma are added 2 volumes of 10 per cent. trichloroacetic acid. The mixture is filtered after standing for 10 to 20 minutes, and 6 ml. of the filtrate are transferred to a 15-ml. conical centrifuge tube. To remove organic and inorganic phosphates, the solution is mixed with 0.5 ml. of 2.5 per cent. aluminium chloride (hexahydrate) solution and 0.5 ml. of ammonia solution (prepared by diluting 20 ml. of ammonia of sp.gr. 0.91 to 100 ml. and adding 5 ml. of 0.02 per cent. methyl red solution). The solution, which should be distinctly alkaline, is allowed to stand until the aluminium hydroxide is flocculated; it is then centrifuged for 5 minutes. Into a second centrifuge tube with a narrow end are introduced 3.5 ml. of the centrifugate, and the solution is neutralised by adding about 1 ml. of freshly prepared 5 per cent. trichloroacetic acid solution in anhydrous acetone. A further 2 ml. of the trichloroacetic acid solution is then added, followed by 6 ml. of a freshly prepared 1.5 per cent. solution of benzidine in anhydrous acetone. After thorough mixing the solution is allowed to stand at 0° C. for 2 hours, and then centrifuged for 15 minutes. The supernatant liquid is decanted, and the centrifuge tube is allowed to drain. The precipitate is washed with 10 ml. of acetone containing 5 per cent. of trichloroacetic acid, the liquid is decanted and the tube drained as before, and the precipitate is dissolved in 2 ml. of warm 0.2 N

hydrochloric acid. The solution is transferred to a 25-ml. graduated tube with the aid of 3 or 4 small portions of water, so that the final volume is 6 to 8 ml. The benzidine solution is treated successively with the following reagents, the tube being thoroughly shaken after each addition: (a) 0.5 ml. of 1 per cent. gum arabic solution (treated before use with a few drops of bromine, the excess of which is expelled by boiling), (b) 1 ml. of phosphotungstomolybdic acid* (Folin and Ciocalteu, *J. Biol. Chem.*, 1927, 73, 627), (c) (after 3 minutes) 2.5 ml. of 16 per cent. sodium carbonate solution, and finally after an interval of at least 10 minutes, to allow the alkali to destroy the excess of reagent, (d) 2 ml. of recently prepared 2 per cent. sodium sulphite solution. This converts the indefinite green colour of the reaction mixture into a stable blue-grey colour; after a few minutes the solution is made up to 20 ml., and the colour is compared with that given by known amounts of benzidine hydrochloride treated in a similar manner. For greater accuracy, the colour may be measured in a Pulfrich photometer with filter S72. The result indicates the amount of *inorganic sulphate* present. To estimate the *total sulphate*, 6 ml. of blood filtrate and 0.5 ml. of *N* hydrochloric acid are evaporated to dryness in a small beaker, the residue is dissolved in 6 ml. of water, and 0.5 ml. of 2.5 per cent. aluminium chloride solution and 0.5 ml. of the ammonia and methyl red solution are added. After centrifuging, 3.5 ml. of the supernatant liquid are treated as described above. The *total sulphur* is estimated by digesting 3 ml. of blood filtrate with 3 ml. of pure conc. nitric acid in a 100-ml. Kjeldahl flask until the liquid turns brown. A few drops of perhydrol are added, and the heating is continued until a white ash remains. This is dissolved in 6 ml. of water and treated with aluminium chloride solution and ammonia, as described above.

Sulphur in urine.—One ml. of urine of normal concentration is transferred to a 50-ml. volumetric flask, and 4 ml. of 0.4 per cent. uranyl acetate solution are added to remove phosphates. After shaking, the flask is allowed to stand for at least 5 minutes, and the solution is then diluted to a definite volume and filtered. To estimate *inorganic sulphate*, 1 ml. (equivalent to 0.02 ml. of urine) is treated with 2 ml. of 10 per cent. trichloroacetic acid solution, and the estimation is continued as for blood. *Total sulphate* is determined on 0.5 ml. of the filtrate; this is treated with 2 to 3 ml. of water and 0.5 ml. of *N* hydrochloric acid and further treated as described for blood. The *total sulphur* is estimated by incinerating 1 ml. of the filtrate with nitric acid and perhydrol as described above.

The colour developed is strictly proportional to the amount of benzidine present. Inorganic sulphate, added to blood and urine, was recovered with an error not exceeding 4 per cent.

F. A. R.

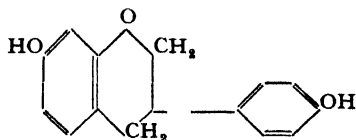
Urothione, a Yellow Sulphur-containing Pigment from Human Urine.
W. Koschara. (*Z. physiol. Chem.*, 1940, 263, 78-79).—A lemon-yellow pigment, to which the name urothione has been given, was isolated from human urine in a

* The phosphotungstomolybdic acid reagent is made by mixing 100 g. of sodium tungstate (dihydrate) and 25 g. of sodium molybdate (dihydrate) with 700 ml. of water, adding 50 ml. of 85 per cent. phosphoric acid and 100 ml. of conc. hydrochloric acid to the mixture, and boiling under reflux for 10 hours. At the end of this period 150 g. of lithium sulphate, 50 ml. of water and a few drops of liquid bromine are added, and the solution is boiled for 15 minutes to expel the excess of bromine. It is then cooled, diluted to 1 litre and filtered. The final solution should be free from any green tint and should be kept protected from dust, as organic materials gradually produce slight reduction.

yield of 40 to 80 mg. per 1000 litres. It contains 20 per cent. of sulphur, and has no definite m.p. It is very soluble in water, alcohol and acetone, dissolves readily in acids and alkalis, but is insoluble in ether and chloroform. When its aqueous solution is layered with conc. sulphuric acid, a cherry-red ring is formed at the junction of the two phases. A solution of the new pigment in sulphuric acid exhibits no fluorescence in ultra-violet light, but after oxidation with potassium permanganate, and removal of the excess with hydrogen peroxide, the solution fluoresces olive-green. This test gives a positive result with a dilution of 1 part in 10 millions, and can actually be demonstrated with urine. Urothione has the formula $C_{11}H_{13}O_3N_2S_2$ and yields one molecule of methyl mercaptan on catalytic hydrogenation; it contains one free amino-group (van Slyke) and gives a tetra-acetyl derivative, m.p. $220^\circ C$. The pigment also occurs in the liver and certain other organs of man and cattle, but nothing is known of its physiological significance.

F. A. R.

Colour Reaction for the Detection of the Equol of Mare's Urine. W. Dirscherl. (*Z. physiol. Chem.*, 1940, **264**, 57-63.)—When certain fractions from mare's urine were layered with conc. nitric acid, a red ring appeared at the junction of the two liquids, and on heating similar extracts with an equal volume of 25 per cent. nitric acid, a red precipitate was produced, which dissolved in ether or amyl alcohol to form red solutions. The red precipitate was also soluble in sodium hydroxide solution and in ammonia, and was re-precipitated on addition of acid. The coloured product was not produced by other acids. The substance in the extracts of mare's urine responsible for the reaction was shown to be equol (7,4'-dihydroxyisoflavan),



The reaction, which will detect 0.1 per cent., can be made even more sensitive (0.01 per cent.) by evaporating a drop of the solution with a drop of nitric acid on a microscope slide and examining the resulting solid under the microscope. Other phenols, including the oestrogenic hormones, produced either a yellow colour or no colour at all, and even the closely related substances daidzein (7,4'-dihydroxyisoflavone) and its methyl ether, formononetin, gave a negative result. Many other substances, such as chromans, coumarans, flavanones and xanthenes, were tested, but only equol gave the characteristic coloured precipitate. Whereas α -tocopherol gives a red colour with nitric acid in alcohol solution (Furter and Meyer, *cf. ANALYST*, 1939, **64**, 217) equol gives a yellow colour, and in small amounts does not interfere with the estimation of tocopherol. Conversely, tocopherol does not give a colour in aqueous solution. The equol derivative responsible for the red colour is believed to be a nitro-quinone.

F. A. R.

Studies on the Anti-Grey Hair Vitamin for Silver Foxes. G. Lunde and H. Kringstad. (*Saertrykk av Norsk Pelsdyrblad*, 1939, **13**, 500-505; *Naturwiss.*, 1939, **45**, 755). In previous experiments it was demonstrated that

rats require a vitamin B_x belonging to the vitamin B complex, the absence of which causes the black colour of the fur to change to grey. The same colour change has now been shown to take place in fox cubs deprived of vitamin B_x , the change being especially evident on the snout. The cubs fed on the vitamin B_x -deficient diet (basal diet with marmite and synthetic aneurin) were noticeably more docile than cubs fed on a complete diet. The new vitamin is heat-labile, and is not adsorbed by fuller's earth.

F. A. R.

Combined Ascorbic Acid of Animal Tissue. P. Holtz. (*Z. physiol. Chem.*, 1940, **263**, 187–205.)—Ascorbic acid occurs in animal tissues and body fluids not only in the free state, but also combined with protein. In the usual method of estimation, that is, after removal of protein, only the free ascorbic acid is estimated. The combined ascorbic acid can be estimated by breaking down the protein-vitamin complex, by hydrolysis with hydrochloric acid or by the action of a proteolytic enzyme such as pepsin or papain, or by autolysis. Of these methods, acid hydrolysis is the most satisfactory. The protein precipitate obtained from the tissue extract or body-fluid by treatment with sulphosalicylic acid or trichloroacetic acid is heated for 10 minutes at 100° C. with 0.5 *N* hydrochloric acid in an atmosphere of carbon dioxide. The solution is then made up to a definite volume with sulphosalicylic acid or trichloroacetic acid solution respectively and filtered or centrifuged, and the clear filtrate is used for the titration. Combined ascorbic acid appears to be the reserve form of the vitamin, and is much more stable to oxidation than the free form.

F. A. R.

Tillmann Reaction for Vitamin C in Plant Extracts. C. Gatti and A. Knallinsky. (*Z. physiol. Chem.*, 1940, **263**, 37–40.)—Experiments with guinea-pigs showed that maté leaves had no protective action against scurvy, nor had an infusion prepared from them. On titrating extracts with 2:6-dichlorophenol-indophenol, however, a reduction value equivalent to 15 to 18 mg. of ascorbic acid was obtained. Solutions of medicinal tannin also reduced the indicator, and since maté contains from 4 to 20 per cent. of tannin-like substances, only a portion of which is precipitated by lead acetate or formaldehyde, it seemed possible that these substances in maté were responsible for its reducing action. Maté was soaked for 20 minutes with cold water and filtered, and the extract was titrated at intervals with the indicator. A steady rise in the titre was observed over a number of days, whereas the reducing power of a solution of ascorbic acid steadily decreased. A solution of tannin heated to 95° C. and then titrated with the indicator gave a high reduction value, thereby behaving like hot aqueous extracts of maté and China tea, but differently from ascorbic acid solutions. A value equivalent to 40 mg. of ascorbic acid per 100 g. was obtained with China tea, although this is known not to possess antiscorbutic activity. The tannins of China tea were completely removed by two or three precipitations with lead acetate and sodium sulphate, and the final filtrate did not reduce 2:6-dichlorophenol-indophenol. The tannin-like substances of maté, however, were much more difficult to remove, and some remained even after four precipitations. The following simple method of freeing maté extracts from tannin substances was used:—One g. of hide powder was added to each 10 ml. of infusion, and, after standing for three hours, the

mixture was treated with lead acetate and filtered, and the precipitation with lead acetate was repeated. The filtrate gave a negative reaction for tannins and did not reduce the indicator. Thus the Tillmann reaction is not applicable to the estimation of ascorbic acid in plant extracts, such as tea and maté, which contain tannins or allied substances. These must be removed before the titration is carried out.

F. A. R.

Colorimetric Determination of Tocopherol (Vitamin E). IV. Quantitative Determination of Tocopherol in Oils after Saponification. A. Emmerle. (*Rec. trav. chim. Pays-Bas*, 1940, **59**, 246-248.)—The stability of tocopherol towards methyl alcoholic potassium hydroxide solution is affected by the amount and concentration of the alkali solution used, and by the temperature and the time of heating. The conditions for saponifying wheat-germ oil and olive oil to produce the minimum destruction of tocopherol were determined. It was found that saponification was complete when 1 g. of wheat-germ oil was heated with 2 ml. of 2 *N* methyl alcoholic potassium hydroxide solution at 72° to 74° C. for 10 minutes, and that the maximum amount of tocopherol was thereafter extracted. The saponifications were carried out in test-tubes (16 × 140 mm.), fitted with small reflux condensers. After saponification 8 ml. of absolute methyl alcohol and 10 ml. of water were added, and the mixture was extracted with three 50-ml. portions of peroxide-free ether. The combined ethereal extracts were washed with dilute alkali and water, dried over pure sodium sulphate and evaporated in an atmosphere of carbon dioxide. The residue was dissolved in a suitable volume of ethyl alcohol or benzene, and the amount of tocopherol was measured by the colour reaction previously described (*cf.* ANALYST, 1939, **64**, 216, 446, 837). It should be noted that this reaction should be carried out in very subdued daylight or in very weak artificial light. Tocopherol added to either wheat-germ oil or olive oil was recovered by this method to the extent of 98 per cent.

F. A. R.

Bacteriological

Bacteria from Chlorinated Waters. M. Levine, P. Carpenter and J. M. Coblentz. (*J. Amer. Water Works Ass.*, 1939, **31**, 1511-23; *Bull. Hygiene*, 1940, **15**, 129-130.)—Bacteriological examinations of the chlorinated water from Lake Michigan yielded 282 strains of the coli-aerogenes group—67 in the winter and 215 in the summer months, the residual chlorine ranging from 0.2 to 0.4 p.p.m. Of these strains, 28 were *B. coli*, 85 *B. aerogenes* and 83 intermediates. Many were obtained from presumptives showing poor gas formation. The question of the sanitary significance of these organisms is of great importance. The resistance of suspensions of cultures of these organisms to chlorine was tested. Suspensions were made from agar slope cultures and adjusted to about a million organisms per ml., chlorine was added to make a concentration of 0.4 p.p.m., and the number of surviving organisms and the residual chlorine (usually 0.2 p.p. 10%) were determined after 20 minutes. None of the winter strains, and only a few of the summer strains, showed any resistance to chlorine. The authors conclude that there is no inherent resistant character of individual organisms, but only resistance

associated with clump formation or protection by some constituent or character of the water. As already stated, 196 (28 + 85 + 83) of the 282 strains were of the coli-aerogenes group; the remainder were non-lactose fermenters and related to the typhoid-paratyphoid-dysentery group, although not absolutely agreeing with the cultural characteristics of these pathogens. One typhoid-like strain agglutinated with typhoid serum at a titre of 1:1000. The survival of these bacteria may indicate the possible survival of pathogenic members of the same genus. It appears therefore that a residual chlorine-content of 0.2 to 0.4 p.p.m. is not an absolute guarantee of effective sterilisation under all conditions.

D. R. W.

Stability of Bacteria in Relation to pH. J. G. Baumgartner and G. G. Knock. (*J. Soc. Chem. Ind.*, 1940, 59, 53-56.)—The five bacterial species *Esch. coli*, *Esch. cloacae*, *Proteus vulgaris*, *Staph. aureus* and a *Lactobacillus* strain, regarded as protein systems, were used to ascertain the optimum pH with regard to (a) heat resistance, (b) precipitation with ethyl alcohol, and (c) flocculation as measured by opacity of the bacterial suspension. Except with the *Lactobacillus* sp., which was grown for 4 days on wort-agar, the suspensions were obtained from 24-hour agar cultures, and the organisms were removed, washed and suspended in water, shaken for 5 minutes, and filtered through cotton-wool pulp. McIlvaine's phosphate-citric acid buffer was used, diluted 1 in 2½, and 100-ml. quantities were prepared for each pH value from 3.0 to 8.0. (a) The heat treatment was carried out by adding 1 ml. of the suitably diluted aqueous suspensions of the bacteria to 25 ml. of the buffer solutions, and, after mixing, 3 ml. of each suspension were heated in thin-walled tubes in a water-bath. After rapid cooling in water 1 ml. of the suspension and 1 ml. of a 1 per 1000 dilution were plated. (b) For the alcohol treatment 5 ml. of absolute alcohol were added to 20 ml. amounts of buffer solution, adjustments of the pH were made when necessary, and the pH values were determined electrometrically at the end of the experiment. The bacteria were added to the alcohol-buffer mixture and held at 22° C., and at the end of the exposure 1-ml. amounts were placed in 19 ml. of sterile water, and plates were prepared. (c) To test the stability in suspension, thick suspensions of the bacteria were made in the buffer solution, 20-ml. amounts being taken in standard tubes (6 × ¾ in.), which were held at 4° C. for 24 hours, and the degree of opacity and sedimentation were estimated visually, *i.e.* the point at which opacity was maximal and sedimentation minimal. In each set of tests excellent agreement between the five bacterial species was obtained with regard to the optimum pH, so that at this point for a given bacterial suspension a common factor in stability appears to be operating, and this is believed to be hydration of the bacterial protein. Three tables are given showing details of experiments. It has not yet been ascertained why different bacterial species have a critical pH value ranging, in the five species examined, from 5.2 to 7.2.

D. G. H.

Agricultural

Determination of the Size Distribution of Soil Clods and Crumbs. E. W. Russell and R. V. Tamhane. (*J. Agric. Sci.*, 1940, 30, 210-234.)—A method of simple sieving will give the required information, provided that the

soil is not too wet; if it is, individual clods smaller than 3 mm. will stick together on the 3-mm. sieve. The method and technique should be varied according to the information required. If many of the crumbs are larger than 0.5 mm., a water-sieving method is essential. For general purposes it is desirable, when possible, to use a very slow or a vacuum-wetting technique and a very rapid wetting technique, such as wetting the soil by immersion in water. When most of the water-stable crumbs are smaller than about 0.1 mm., the hydrometer method will be adequate unless separated crumb fractions are required; elutriation is then essential. For general purposes the use of air-dry soil is recommended. Very lumpy soils must be sieved before it is possible to take a 100-g. sample; pre-sieving on a $\frac{1}{8}$ -inch sieve facilitates good sampling. The distribution curves for the clods and the residual soil should be determined separately, and that for the whole soil calculated from them. It has been found possible to separate the crumb fractions of Rothamsted soils into crumbs and sand particles by dispersing them in a mixture of bromoform and carbon tetrachloride of the correct density; the crumbs float, while the sand particles sink. There is always some loss of soil in this method, but it is usually less than 5 per cent. A rapid means of overcoming the difficulty of distinguishing between crumbs and sand particles is to determine the proportion of the total base exchange capacity of the soil present in each crumb fraction. For this purpose the rapid method of Schofield (*J. Agric. Sci.*, 1933, **23**, 252) is recommended. The degree of aggregation of a soil is usually defined as that proportion of the soil or of some soil component which is in crumbs larger than a certain size d . There is no optimum choice for the crumb size of d ; the value $d = 0.5$ mm. has usually been chosen at Rothamsted as affording the most information about the different soils studied.

Distribution of Manganese in the Pea Seed in Relation to Marsh Spot.
H. H. Glasscock and R. L. Wain. (*J. Agric. Sci.*, 1940, **30**, 132-140.)—The disease known as marsh spot is confined to the seed of the culinary pea, and its occurrence is noted yearly in certain districts in this country and elsewhere in Europe, especially Holland. The cells of the affected areas die and form a lesion, involving the cotyledons, which show spots having a water-soaked appearance. Previous investigators have failed to associate the cause of the disease with fungi, bacteria or a virus. The present work indicates that the cause is a deficiency of available manganese in the soil. For the determination of manganese, the colorimetric method of Willard and Greathouse (*J. Amer. Chem. Soc.*, 1917, **39**, 2366; *Abst.*, *ANALYST*, 1918, **43**, 44) was used, and for whole peas the procedure of Richards (*ANALYST*, 1930, **55**, 554) was adopted. In the diseased peas the highest amount of manganese (5 p.p.m.) was found in the peripheral tissues of the cotyledons followed by the germ (3 p.p.m.) and seed-coat (2 p.p.m.). The figures for a healthy sample were 15 p.p.m. in the germ, 11 p.p.m. in the outer tissues of the cotyledon, and 4 p.p.m. in the seed coat. A given weight of small peas contained less manganese than the same weight of large peas selected from the diseased sample, whereas the reverse was found for groups of similar size from the healthy sample. Thus in the diseased large peas the amounts of manganese ranged from 4.0 to 4.3 p.p.m., and in the small peas from 3.3 to 3.5 p.p.m., whilst in the healthy

peas the range was from 8.3 to 9.3 p.p.m. (average 8.7) for large seeds and from 8.4 to 9.9 p.p.m. (average 9.3) for small seeds. It is suggested that migration of cell contents from the necrotic tissue of diseased peas may partly account for the differences in the manganese-contents of healthy and diseased peas.

Determination of Pyrethrin I. The Seil Colour Reaction in the Mercury Reduction Method. C. S. Sherman and R. Herzog. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 136–137.)—Wilcoxon's method (*Contrib. Boyce Thompson Inst.*, 1936, 8, 175–181) and Holaday's revised method (*Ind. Eng. Chem., Anal. Ed.*, 1938, 10, 5) are based on the interaction of pyrethrin I acid and Denigès' reagent (acid mercuric sulphate). Both methods give erratic results, owing to the presence of substances other than pyrethrin I acid which interfere with the titration of the resulting mercurous chloride with potassium iodate. To obtain readable end-points, excess of filter-paper in the titration mixture must be avoided. The colours accompanying the reaction (Seil's colour reaction) appear to be due to the formation of a colloidal dispersion of metallic mercury, or of some mercury compound, which on standing undergoes spontaneous successive increases in particle size until ultimately a coarse blue suspension results. Sodium chloride was found to be the best precipitant for complete conversion of the mercurous ion into calomel and for elimination of fine suspended material. Provided that the mixture is centrifuged for a short time after addition of the sodium chloride, it is not necessary to allow more than 15 minutes between adding the Denigès reagent and the sodium chloride.

D. G. H.

Organic

o-Nitrobenzazide as a Reagent for the Identification of Phenols.

P. P. T. Sah and W.-H. Yin. (*Rec. Trav. Chim. Pays-Bas*, 1940, 59, 238–245.)—In boiling ligroin solution *o*-nitrobenzazide decomposes to give gaseous nitrogen and *o*-nitrophenyl isocyanate, the latter reacting with phenols to form crystalline *o*-nitrophenyl urethanes, which can be used to identify the phenols. The azide (0.5 g.) is mixed with a molecular equivalent of the phenol in a small Erlenmeyer flask, covered with 5 ml. of anhydrous ligroin, and heated on a sand-bath under reflux for 4 to 5 hours. With ortho-substituted phenols 2 drops of freshly distilled dimethylaniline are added as catalyst. After cooling and standing overnight the urethanes are filtered off under suction and then recrystallised from suitable solvents (usually ligroin, sometimes ethyl acetate, and occasionally a mixture of the two). Tables show the melting-points, crystalline forms and nitrogen-contents of the *o*-nitrophenyl urethanes prepared from 28 phenols, and also a comparison of the melting-points of the *o*-, *m*-, and *p*-nitrophenyl urethanes from the same phenols. In general, the *para*-isomers have the highest melting-points and the lowest solubilities, and the *ortho*-isomers the lowest melting-points and highest solubilities, the values for the *meta*-isomers coming between these. The exceptions noted are (1) 2,4,6-trichlorophenol and 2,4,6-tribromophenol, which yield *m*-nitrophenyl urethanes with slightly higher melting-points than those of the *p*-nitro isomers; (2) hydroquinone monomethyl ester, the *o*-nitrophenyl urethane

from which has a higher m.p. than the *m*-nitro isomer, though lower than that of the *p*-nitro isomer; (3) *o*-iodophenol, the *o*-nitrophenyl urethane from which has a higher m.p. than those of the *m*-nitro and *p*-nitro isomers. E. M. P.

***p*-Nitrobenzazide and *p*-Nitrophenyl Isocyanate as Reagents for the Identification of Amines.** P. P. T. Sah. (*Rec. Trav. Chim. Pays-Bas*, 1940, **59**, 231–237.)—In boiling anhydrous toluene solution *p*-nitrobenzazide decomposes to form *p*-nitrophenyl isocyanate; this reacts with amines to yield crystalline *p*-nitrophenyl ureas which can be used to identify the amines. The nitrobenzazide (0.19 g.) or an equivalent quantity of *p*-nitrophenyl isocyanate (prepared by refluxing *p*-nitrobenzazide in absolutely dry toluene and in complete absence of moisture, distilling under vacuum, and collecting and recrystallising from carbon tetrachloride the portion boiling at 160–165° C. at a pressure of 18–20 mm.) is dissolved in 5 ml. of anhydrous toluene in a small Erlenmeyer flask. Slightly more than a molecular equivalent of the amine is dissolved in 5 ml. of dry toluene, and the solutions are mixed and heated under reflux on a sand-bath for 4 hours. After standing overnight the *p*-nitrophenyl urea is filtered off under suction and recrystallised from a suitable solvent (usually 95 per cent. ethyl alcohol, sometimes acetone, ethyl acetate, ligroin, or benzene), and the melting-point is determined. Tables show the melting-points, crystalline forms and nitrogen-contents of the *p*-nitrophenyl ureas from 50 amines. In general, primary amines give the best results; the ureas from secondary amines are rather soluble in toluene, so that concentration of the solvent or the use of ligroin is necessary to isolate the pure urea; amides and anilides also react, but the yields are decidedly less and the products are more difficult to purify. For some of the ureas it was necessary to use as the melting-bath liquid the alkyl esters of phthalic acid (Sah and Kao, *J. Chinese Chem. Soc.*, 1937, **5**, 86–88; Brown, *J. Chem. Education*, 1937, **14**, 380) with which it is possible to determine melting-points as high as 320° C.

E. M. P.

Little-known Reaction for Benzoic Acid. N. Schoorl. (*Pharm. Weekblad*, 1940, **77**, 425–427.)—The method is a modification of the reactions of Guerbet (*ANALYST*, 1920, **45**, 334; 1921, **46**, 11) and of Guignes (*Bull. Soc. Pharmacol.*, 1928, **35**, 292), in which the benzoic acid is converted into *m*-nitrobenzoic acid, which is reduced to *m*-aminobenzoic acid, and this is diazotised and coupled with β -naphthol to produce a coloured compound. The sample is nitrated with 1 drop of fuming nitric acid (sp.gr. 1.50), and the mixture is evaporated to dryness on the water-bath. The yellow residue is washed into a test-tube with a few drops of 4 *N* sodium hydroxide solution, and reduced by addition of a few drops of a 10 per cent. solution of stannous chloride in 4 *N* hydrochloric acid. The solution is warmed and then cooled, a piece of aluminium is added and cooling is continued; the object of this is to precipitate the tin, a large excess of which interferes with the subsequent formation of the colour. A few drops of 1 per cent. sodium nitrite solution are then added, and the solution is shaken and made alkaline with an ammoniacal solution of β -naphthol. In presence of benzoic acid a bright red colour develops; sensitiveness, 0.1 mg. Cinnamic acid produces the same colour, since the nitric acid oxidises it to benzoic acid. Salicylic acid and *p*-hydroxybenzoic acid give

a dark brown and a dark-red brown precipitate, respectively. If, however, it is desired to test for benzoic acid in presence of these, they may be oxidised with an alkaline solution of potassium permanganate, which does not affect the benzoic acid. If the solution is then acidified with sulphuric acid, the benzoic acid may be extracted with ether. This method, however, does not distinguish benzoic acid from cinnamic acid, since the two acids behave similarly (see also Pesetz, *Pharm. Weekblad*, 1940, 77, 403). J. G.

Essential Oil of Massoi Bark. T. H. Meijer. (*Rec. Trav. Chim. Pays-Bas*, 1940, 59, 191–201.)—The principal oil-containing barks of Dutch New Guinea, Massoi and Lawang, may be distinguished by the fact that Lawang oil contains eugenol and therefore gives a greenish colour with aqueous ferric chloride in alcoholic solution, whereas Massoi oil does not. Abe (*J. Chem. Soc. Japan*, 1937, 58, 246–251), who was the first to investigate real Massoi oil, found that it had the following characteristics: n_D^{20} 1.4721, sp.gr. at 20°/20° C. 0.9822, and optical rotation $\alpha_D = -87.01^\circ$. The author obtained the following values for authentic samples of the oil:

| | | | | | | |
|-----------------------------|----|----|--------|--------|--------|--------|
| n_D^{20} | .. | .. | 1.4770 | 1.4726 | 1.4717 | 1.4749 |
| Sp.gr. at 27.5°/4° C. | .. | .. | 0.9821 | 0.9643 | 0.9695 | 0.9782 |
| Optical rotation α_D | = | | -94° | -64° | -74° | -106° |

The yield of essential oil varied between 1.7 and 2.4 per cent. The constants of two samples of genuine Lawang oil were: n_D^{20} , 1.534 and 1.5350; sp.gr. at 15°/15° C., 1.060 and 1.0559; rotation, -0.9° and -0.8° ; eugenol, 60 and 94 per cent.

Massoi oil contains a lactone which on hydrogenation takes up one molecule of hydrogen; oxidation of the dihydrolactone with potassium dichromate and sulphuric acid yields valeric, caproic, succinic, glutaric and δ -keto-capric acids. Caproic acid is also obtained by oxidising Massoi oil lactone with permanganate or with dichromate and sulphuric acid; with the latter reagents fumaric acid is also formed. E. M. P.

(Note.—Oil from the Massoi bark of commerce has been found to correspond more closely with the Lawang oil as described above.—EDITOR.)

Determination of Hydrocarbons in Unsaponifiable Matter. J. Grossfeld (*Z. Unters. Lebensm.*, 1939, 78, 273–285.)—On shaking 3 ml. of potassium hydroxide solution (sp.gr. 1.50), 20 ml. of 96 per cent. alcohol, 20 ml. of water and 50 ml. of benzine (b.p. 60–70° C.) with a weighed quantity of paraffin (paraffin-wax or mineral oil), and, after separation of the phases, evaporating 25 ml. of the benzine solution, recovery of the paraffin is quantitative. With cholesterol there is a 98 per cent. recovery. If, however, palmitic acid is added, paraffin and cholesterol behave differently. Under these conditions the proportion of paraffin (calculated on 50 ml. of benzine) becomes 109 per cent., and that of the cholesterol only 26 per cent. The apparent high yield of paraffin is due to the diffusion of benzine into the soap solution; this amounts to 7 ml., so that the concentration of paraffin in the residual 43 ml. is increased. A new analytical constant, the hydrocarbon value (HCV),* has been based on this difference. A weighed quantity of the unsaponifiable matter (e.g. 0.5 g.) is mixed with 5 g. of a fatty acid (conveniently palmitic or

* HCV is used here to represent KWZ (Kohlenwasserstoffzahl) in the German original paper.

oleic acid), 20 ml. of 96 per cent. alcohol, 3 ml. of potassium hydroxide solution (sp.gr. 1.50) and a few granules of pumice, and boiled for 5 minutes beneath a reflux condenser. The mixture is then cooled to about 38° C. if palmitic acid was used or to about 15° C. for oleic acid, and gently mixed with 50 ml. of benzine (b.p. 60°–70° C.). After further cooling if palmitic acid was used, 20 ml. of water are added, and the flask is closed with a rubber stopper, shaken and allowed to stand overnight. Twenty-five ml. of the benzine solution are then transferred to an Erlenmeyer flask by means of a pipette, the benzine is distilled off, and the residue is dried at 105° to 110° C. and weighed. A control test is made simultaneously with the reagents alone, and the amount of residue obtained is deducted from the residue containing the unsaponifiable matter. The following constants (P.HCV = palmitate hydrocarbon value; O.HCV = oleate hydrocarbon value) were determined:

| | | Paraffin | Sterol | Cetyl alcohol |
|-------|----|----------|--------|---------------|
| P.HCV | .. | 109 | 26 | 39 |
| O.HCV | .. | 107 | 27 | 43 |

For calculating the amount of paraffin in presence of sterol only, the formula is:

$$x = 1.21 (P.HCV - 26) \text{ or } 1.25 (O.HCV - 27).$$

If only cetyl alcohol is present with paraffin,

$$x = 1.43 (P.HCV - 39) \text{ or } 1.56 (O.HCV - 43).$$

For mixtures of cetyl alcohol and sterol the amount of the former is calculated by the formula: $C = 7.7 (P.HCV - 26) \text{ or } 6.25 (O.HCV - 27)$. When the unsaponifiable matter contains the three components, one must be determined separately, e.g. sterols by precipitation with digitonin. The two others can then be calculated from the hydrocarbon value. For example, in a test experiment in which a mixture of 0.120 g. of paraffin, 0.210 of cetyl alcohol and 0.155 g. of cholesterol was taken, the amount of paraffin found was 0.119 g.

D. A.

Application of Rational Analysis to some Typical South African Coals.

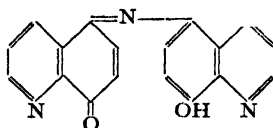
G. K. Morrison. (*J. Chem. Metall. Mining Soc. S. Africa*, 1939, 40, 201–212.)—The main constituents of coal from the point of view of rational analysis are the hydrocarbons and resins, the organised plant entities (spore exines, cuticles, etc.), the ulmins, and fusain. Francis and Wheeler (*J. Chem. Soc.*, 1928, 2967) have shown, however, that, since no correlation exists between the proportion of these constituents and the maturity and rank of a coal, no system of classification can be derived from them; more information is obtainable from the reactivity of the ulmins towards oxygen. In determining the reactivity the hydrocarbons and resins are first removed by extracting the sieved coal (passing 60 and retained on 120 I.M.M. sieve) with boiling pyridine for 8 hours. If most of the solvent is then removed from the extract, and the residue is acidified, thoroughly dried and extracted with dry ether, the residue, after evaporation of the ether, gives the hydrocarbons and resins. Although this extraction process is necessary and satisfactory as a preliminary to the determination of ulmin activity, it is unsatisfactory as a method for the determination of hydrocarbons and resins, owing to the dependence of the results on the purity of the pyridine and to

the slow removal of the hydrocarbons and resins from the pyridine extract. The (resistant residue + fusain) is obtained by boiling 0.5 g. of the sieved extracted coal (passing 120 and retained on 150 I.M.M. sieve) for 7 hours under a reflux condenser with 50 ml. of oxidising solutions (mixtures of potassium chlorate and *N* nitric acid) of different strengths. With the aid of a microscopical examination of the residue, the oxidising agent can be chosen so that the ulmins are dissolved out, whilst the resistant plant entities are unattacked. The weight of residue after oxidation (*P* g.) gives therefore, the amount of resistant residue plus the insoluble ash in the coal. With the four South African coals examined, however, there were indications that portions of the resistant residue are attacked before all the ulmins have been rendered soluble, or that a modified form of ulmin is present, the oxidisability of which overlaps that of the true ulmin and that of the resistant residue. The method therefore is only approximate as a means of classification, and it is usually preferable to determine the resistant residue in the clarain (*cf.* Francis and Wheeler, *ANALYST*, 1931, **56**, 333, 420); here again, however, this presented difficulties with the South African coals because of their small clarain contents. An alternative measure of the resistance of ulmins to oxidation, and consequently of the maturity of a coal, is Francis and Wheeler's "reactivity index." This is the percentage of the ulmins rendered soluble by oxidation of 0.5 g. of the coal (after extraction with pyridine, as described) with a solution of 0.9 g. of potassium chlorate in 50 ml. of *N* hydrochloric acid. The reaction is carried out in a pressure-bottle, which is immersed in boiling water for 7 hours and then allowed to cool over-night, the oxidised ulmins being dissolved out with *N* potassium hydroxide solution and the insoluble residue dried and weighed (*R* g.). If the weights of ash from 0.5 g. of coal after extraction with pyridine and from the above *R* g. of residue are *A* and *A'*, respectively, then the reactivity index is given by $100 [(0.5 - A) - (R - A')] / [(0.5 - A) - (P - A')]$. An examination of the experimental accuracy of the above procedures is presented for the four South African coals, with particular reference to the effects of the purity of the pyridine; it is concluded that the resulting variations in the results are no greater than those obtained by repeated determinations on the same sample, but that the differences obtained by repeated tests on the same sample are so large that many determinations must be made in order to calculate the reactivity index with any degree of certainty. In view of the limitations of the method, Francis (*Fuel*, 1932, **11**, 171; 1933, **12**, 128) and Heathcoat (*id.*, 1933, **12**, 4) have suggested that an alkaline permanganate solution should prove more satisfactory than the acid oxidising agents; 0.5 g. of the extracted sample is therefore ground to pass a 120 I.M.M. sieve but be retained on a 150 I.M.M. sieve, and oxidised by the action of 50 ml. of *N* sodium hydroxide and 200 ml. of *N* potassium permanganate solutions for 1 hour in a boiling water-bath. Then the permanganate number is the number of ml. of *N* potassium permanganate solution utilised by 0.5 g. of ash-free extracted coal; it is expressed in terms of the percentage of ulmins rendered soluble or of the amount of oxidising agent used up. It is concluded that more confidence may be placed in this value, as the experimental error is small, and there is little variation in the results obtained from the same coal by different workers. According to Francis (*loc. cit.*) oxidation of coal by the atmosphere at low temperatures, or by

Hofmeister's reagent, is effectively complete only when the ulmins have been rendered soluble in alkalis, attack being confined to the external groupings of the ulmin molecule. Since, however, alkaline permanganate attacks the nuclear structure also, it provides a measure of the reactivity of coal to a stage further in the direction of complete combustion. The relative merits of the two methods depend, therefore, on the property of the coal which it is desired to investigate. Thus the reactivity index may be expected to correlate well with properties corresponding with the initial stages of combustion (*e.g.* spontaneous combustion or the first stages of destructive distillation), whilst the permanganate number should give a better indication of processes involving the complete breakdown of the molecule (*e.g.* hydrogenation, chlorination and high-temperature distillation). J. G.

Inorganic

Indo-Oxine, a new Precipitant for Metals. R. Berg and E. Becker. (*Z. anal. Chem.*, 1940, 119, 81-90.)—The reagent is quinolinequinone (5, 8)-[8-hydroxyquinolyl(5)-imide]-5, a reddish-brown crystalline powder of m.p. 253°–254° C., soluble in mineral or glacial acetic acid to form a red solution which changes to green with excess of alkali. In dilute solutions a colour change takes



place at pH 6 to 8. Indo-oxine is a serviceable indicator for the titration of very dilute (*e.g.* 0.01 *N*) acid or alkali, but carbon dioxide interferes and must be expelled by boiling. In acetic acid or neutral solutions indo-oxine forms a sparingly soluble bluish-green silver salt, and it can therefore be used as an indicator for the titration of halogens. It also forms blue or bluish-green precipitates with a number of other metals in acetic acid or ammoniacal solution, and is particularly suitable for the micro-determination of copper, nickel and mercury. *Copper*.—A neutral solution, containing from 0.2 to 1.0 mg. of copper, is treated with 5 ml. of 2 *N* acetic acid and 5 ml. of conc. sodium acetate solution, diluted to 50–60 ml. and heated to 60°–70° C., after which it is titrated with an alcoholic 0.05 per cent. solution of indo-oxine. Since the blue colour of the precipitate masks the red colour of the liquid, it is necessary after each addition of the reagent to filter a few drops through a quantitative micro-filter. *Nickel* and *mercury* are determined in the same way, but the mercury solution must be free from halogens, which form soluble complex compounds with the reagent. Thus minute quantities of copper may be titrated in presence of mercury by nearly neutralising the solution (containing 0.2 to 1 mg. of copper and 0.25 to 0.75 g. of mercury), adding 5 ml. of 2 *N* acetic acid, 5 ml. of conc. sodium acetate solution and 5 g. of sodium chloride, heating the mixture to 60°–70° C. and titrating it as described above. W. R. S.

Partial Separation of Copper from Small Amounts of Arsenic. B. Park. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 97–98.)—The sample of copper (10 g.) is dissolved in a mixture of 100 ml. of water, 20 ml. of conc. nitric acid and 20 ml.

of conc. sulphuric acid. Nitrogen oxides are removed by heating, 10 g. of tartaric acid are added, the liquid is diluted to 350 ml., and the bulk of the copper is removed by electrolysis. Arsenic may be determined in the solution by any convenient method.

S. G. C.

Stability of Peroxidised Titanium Solutions. G. H. Ayres and E. M. Vienneau. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 96.)—Colorimetric studies showed that the yellow colour developed by titanium and hydrogen peroxide in dilute sulphuric acid remained unchanged for 2 years, indicating great stability of the colour. The use of artificial colour standards is therefore unnecessary.

S. G. C.

New Method for the Removal of Nitric Acid. E. Müller and C. Burchard. (*Z. physiol. Chem.*, 1940, 263, 47–48.)—The removal of large quantities of nitric acid is best accomplished by conversion into barium nitrate, which is sparingly soluble in methyl or ethyl alcohol. Baryta is added to the solution, and carbon dioxide is passed through it to precipitate the excess. After separation of the barium carbonate, the filtrate is concentrated to a small bulk and four volumes of methyl alcohol are added. Most of the barium nitrate is precipitated and is filtered off after standing overnight; the filtrate is then distilled under reduced pressure to remove the alcohol. The rest of the nitric acid is removed by the following procedure, which is also recommended when only small amounts of nitrates are present in a solution:—A solution of 15 g. of nitron in 100 ml. of 5 per cent. sulphuric acid and 500 ml. of water is added until no further precipitate forms. The insoluble nitron nitrate is immediately filtered off, and the filtrate is treated with aqueous picric acid solution, a large excess being avoided. The precipitate of nitron picrate is filtered off, the filtrate is acidified with sulphuric acid, and the picric acid is extracted with ether. After removal of the sulphuric acid, a solution of the free base remains.

F. A. R.

Microchemical

Rapid Method for the Accurate Estimation of Minute Quantities of Arsenic in Biological Material. J. Bodnár, Ö. Szép and V. Cielešky. (*Z. physiol. Chem.*, 1940, 264, 1–22.)—Gangl and Sánchez (*Z. anal. Chem.*, 1934, 98, 81; Abst., *ANALYST*, 1934, 59, 716) introduced three improvements into the ordinary Marsh test. The first of these consisted in the use of zinc with a large surface-area, the second in the use of a quartz spiral instead of a straight tube for producing the arsenic mirror, and the third in dissolving the arsenic in iodine chloride solution and titrating the liberated iodine with potassium iodate solution after addition of potassium cyanide. Several further refinements have now been incorporated in this method, enabling the estimation of 0.3 to 5γ of arsenic to be made with a high degree of accuracy. The method comprises three separate stages. For the first stage, a 100-ml. conical flask fitted with a stopper carrying two tap-funnels and an air condenser is used. The minced tissue (from 10 to 15 g. according to the arsenic-content), or the dry solids obtained from such tissue or body fluid (2 to 3 g.), are weighed into the flask and thoroughly mixed by stirring

ABSTRACTS OF CHEMICAL PAPERS

with 1 ml. of 50 per cent. sodium chlorate solution. The flask is then stoppered, and 4 ml. of fuming hydrochloric acid (sp.gr. 1.19) are run into it from one of the tap-funnels. The flask is immersed in a boiling water-bath, and after a few minutes a further ml. of acid is run in with constant shaking, followed by 2 ml., which are added dropwise. When the reaction-mixture begins to froth, the flask is removed from the water-bath, and 1 ml. of 50 per cent. sodium chlorate solution is added from the second funnel, and another 1 ml. of acid simultaneously from the other tap-funnel. After a second period of heating, the decomposition is complete and the solution is allowed to cool and then diluted with a little dilute hydrochloric acid (1 part of conc. acid and 2 parts of water) and warmed to expel residual chlorine. The second stage of the estimation comprises the reduction of the solution and the formation of an arsenic mirror. Spongy tin and hydrochloric acid are used as the reducing agent; unlike zinc and sulphuric acid they do not convert antimony into its hydride. Tervalent arsenic compounds are reduced in the cold, but quinquevalent compounds only on boiling. Consequently, trivalent compounds, if present, must first be oxidised; otherwise arsine may be liberated during the preliminary removal, by hydrogen, of air from the apparatus. Twenty g. of spongy tin* and 6.5 ml. of fuming hydrochloric acid are added to the flask in which the digestion was carried out, and then sufficient water to cover the tin. The flask is closed with a stopper carrying, first, a tube connected (*via* wash-bottles containing potassium permanganate solution and conc. sulphuric acid) with a hydrogen generator and, secondly, a 10-ml. pipette bent at right angles. The other end of the pipette is connected by rubber-tubing with the quartz spiral already referred to (internal diameter 1 mm. at the end connected with the pipette, 0.2 mm. for the rest of the tube), and this in turn is connected with a small tube dipping into water to indicate the rate of flow of the gas. The pipette serves as a condenser and has a length of woollen thread wrapped round it, this being kept moist by a stream of cold water. A similar thread is wrapped round the far end of the quartz spiral where the arsenic mirror is to be formed. The current of hydrogen is turned on and, after about 12 minutes, the spiral is heated with the flame of a fish-tail burner, 8 cm. long and 3 to 4 cm. high. The current of hydrogen is reduced in speed, and the flask is gently heated over an asbestos gauze, the temperature being gradually raised so that boiling begins in 30 to 40 minutes. As hydrogen is evolved by the contents of the flask, the stream of hydrogen is cut off altogether. Boiling is continued for a further 10 minutes, after which both flames are extinguished, and the apparatus is allowed to cool with a current of hydrogen passing through it from the generator. The third stage comprises solution of the arsenic mirror formed on the quartz tube and titration of the arsenic solution with the aid of a micro-burette. Into a 5-ml. titration flask is introduced 0.2 ml. of iodine chloride solution (prepared by dissolving 1.56 g. of potassium iodide and 1 g. of potassium iodate in 50 ml. of water and pouring the solution into 50 ml. of conc. hydrochloric acid. A small

* The spongy tin is prepared by inserting sticks of zinc or aluminium (2 to 4 mm. diameter) in a (diluted) solution of stannous chloride in hydrochloric acid. The precipitated tin is filtered off and thoroughly washed. It is not arsenic-free, however, and is therefore boiled for 1 to 2 hours with conc. hydrochloric acid diluted with an equal quantity of water. The traces of arsenic present are reduced to arsine, which is boiled off, and the undissolved arsenic-free tin is filtered off, washed and dried. The solution can be used for preparing further quantities.

amount of free iodine is removed by adding dilute potassium iodate solution dropwise until a drop of carbon tetrachloride added to the solution is no longer coloured). The narrow end of the quartz tube is immersed in this solution, which is forced in and out of the tube until the mirror is dissolved. The tube is then washed through with 0.06 ml. of iodine chloride solution from a fine pipette, followed by a few drops of dilute hydrochloric acid. Into the flask are then put 0.3 ml. of 10 per cent. potassium cyanide solution and 3 to 4 drops of carbon tetrachloride. The flask is stoppered, vigorously shaken and allowed to stand for 15 minutes. The liberated iodine is titrated with 0.001 *M* potassium iodate solution from a specially designed 0.1-ml. micro-burette graduated in 0.001 ml. The end-point is indicated by the disappearance of colour from the carbon tetrachloride. A blank estimation must be made upon the reagents only, the value obtained being subtracted from the value found for the test solution. The whole operation can be carried out in less than 3 hours, and arsenic added to biological material in amounts varying from 0.3 to 5%, was estimated with a maximum error of +3.7 or -3.4 per cent.

F. A. R.

Micro-Determination of Zinc by the Hydroxyquinoline Method. IV. Separation from Manganese, Iron, Bismuth, Mercury, Arsenic and Antimony. C. Cimerman and P. Wenger. (*Mikrochem.*, 1939, 27, 76-84.)—

I. Separation from manganese salts.—A double precipitation is necessary. The solution, containing about 1 to 3 mg. of zinc in 1.5 ml., is treated in a micro-beaker of Jena glass with a drop of universal indicator, 2 drops of 10 per cent. acetic acid and a few drops of a 40 per cent. solution of sodium acetate until the pH is 5-6. The solution is then heated to boiling, treated with excess (0.1 to 1 ml.) of a freshly prepared 1 per cent. alcoholic solution of 8-hydroxyquinoline, and filtered after 10 to 30 minutes through a filter-stick. The precipitate is washed with double-distilled water and dissolved in 3 ml. of 2 *N* hydrochloric acid and 2 ml. of water. Then 0.5 ml. of 40 per cent. sodium hydroxide solution is added, followed by a 4 per cent. solution until a slight cloudiness appears; this is dissolved in glacial acetic acid, and the solution is treated with about 2 ml. of glacial acetic acid and a solution of 3 g. sodium acetate in water. Water is added to bring the volume to 10 or 20 ml. according to the amount of manganese present. *II. Zinc in presence of iron.*—The solution is treated in a 50-ml. conical flask with 1 ml. of 30 per cent. tartaric acid solution, 2 drops of a 0.1 per cent. alcoholic solution of bromothymol blue and sufficient 8 per cent. sodium hydroxide solution to change the indicator colour. Zinc is precipitated with the reagent at 120° to 130° C. in about 10 ml. of the solution. *III. Zinc in presence of bismuth.*—The precipitation is made in alkaline solution on the same lines as II above. *IV. Zinc in presence of mercury.*—The precipitation is made in alkaline solution as in II, but potassium cyanide is added to prevent precipitation of the mercury 8-hydroxyquinoline compound; as small an excess as possible must be added. *V. Zinc in presence of arsenic and antimony.*—The same procedure is used as in II, the precipitation being made from alkaline solution. The method is the same. It is not possible to effect a micro-analytical separation of zinc from nickel or cobalt by the method. A summary of previous papers on the hydroxyquinoline method is appended.

J. W. M.

Microchemistry of the Rare Earths. G. Beck. (*Mikrochem.*, 1939, 27, 47-51.)—*Determination of scandium and thorium.*—Scandium and thorium alone among the rare earths give precipitates with alizarine-3-sulphonate, difficultly soluble in acetic acid. Very small amounts of scandium and thorium may be separated in a single precipitation from the weakly basic fraction of the rare earths. A 2.5 per cent. solution of sodium alizarine-3-sulphonate is used as reagent, with subsequent addition of 30 per cent. acetic acid. The precipitate is very voluminous and unsuitable for filtration with suction, owing to its slimy nature. It is washed with water, centrifuged, and finally dried at 100° C., and is then in a suitable state for conversion into sulphate by treatment with sulphuric acid containing a little nitric acid. *Reactions of rare earths with tincture of cochineal.*—The rare earths and zirconium compounds yield violet solutions with tincture of cochineal, whereas thorium salts give blue solutions. Only lead and copper salts interfere, as they give a blue precipitate with the reagent. Beryllium, aluminium and gallium salts merely give a pink colour with a red fluorescence in ultra-violet light. Red colours are given by zinc, mercury, cadmium, calcium, magnesium, barium, strontium, nickel, cobalt and manganese salts. The violet solutions of the cerite earths from lanthanum to samarium become reddish-orange on the addition of dilute acetic acid, the original colour of the cochineal tincture being restored. With terbium and erbium earths and zirconium, strong acetic acid is necessary to change the colour to red. The concentration limit for the earth metals is 10 γ in 1 ml. J. W. M.

Physical Methods, Apparatus, etc.

Luminescence Analysis of Rare Elements. M. Servigne. (*Bull. Soc. Chim.*, 1940, 184, 121-132.)—Most of the rare earths, when incorporated in a suitable solid, will form phosphors which give characteristic cathodo- or photoluminescence. At ordinary temperatures the emission spectrum consists of lines or narrow bands capable of precise measurement. The emission spectra of phosphors activated by praseodymium, neodymium, samarium, europium, terbium, dysprosium, holmium, erbium and thulium occur in the visible region, and by gadolinium, lanthanum, and cerium in the ultra-violet; infra-red emission is given by praseodymium, neodymium, samarium, europium and dysprosium. The luminescence of these phosphors is not a simple phenomenon; in addition to the activating element it owes its particular characteristics to several other factors, such as chemical composition of the base material, method of preparation, preliminary heat treatment and final crystalline state of the luminescent solid solution. A further variable is the nature and intensity of the exciting energy. When more than one activator is present, masking effects may occur. The more sensitive activators, Pr, Sa, Eu, can be detected at concentrations of 1 in 100,000. The author considers calcium tungstate to be a suitable base material for activation by rare earths. The calcium tungstate is prepared by heating an intimate mixture of calcium oxide and tungstic oxide at 1100° C. for 2 hours. It should contain less than 1 p.p. 10⁵ of iron and should be free from lead, copper and silver. The best conditions for observing the luminescence spectra are obtained when the phosphor is held at 90° C. The author describes two methods depending on

photo-luminescence. In the first, the source of excitation is a quartz mercury discharge tube (7 mm. diam., 0.5 mm. wall thickness), 30 cm. long and consuming 5 watts. A tube of this type gives 65 per cent. of its energy in the U.V. at 3650 Å and 2537 Å; tungstates are particularly sensitive to the latter radiation. The luminescent substance is placed on a quartz sleeve in a small electric furnace surrounding the tube, and observations are made through a small window in the furnace. In the second method, a mercury discharge tube in the shape of a double U is operated with the substance coated on the inside walls of the tube near the electrodes. This method avoids the necessity for a furnace. The luminescence is photographed on suitable plates. In the infra-red, exposures up to 48 hours may be necessary. Limiting concentrations of various rare earths in calcium tungstate, as determined by these methods, are tabulated. For quantitative estimates the intensities of the spectra are compared photometrically with those of prepared standards. It is claimed that samarium and other rare earths can be determined with a precision of the order of 5 per cent. B. S. C.

Spectrovolumetric Determination of Alkaline Earths and Phosphate.

T. Török. (*Z. anal. Chem.*, 1940, 119, 120–125.)—The non-volatility of the phosphates of the alkaline earths in a Bunsen flame (*ANALYST*, 1939, 64, 383) can be utilised for the volumetric determination of either alkaline earths or phosphate, the technique described in the abstract referred to above being applied. The strongly acid chloride solution of the alkaline-earth metal, containing some pieces of zinc, is titrated with *N* ammonium dihydrogen phosphate solution until the flame placed horizontally above the solution, viewed through a spectroscope, fails to give the characteristic lines of calcium, strontium, or barium. The method is reasonably accurate, and can be used conversely for the titration of acid solutions of phosphate by means of *N* strontium chloride solution. W. R. S.

Low-Temperature Cooling Baths. H. L. Wikoff, B. R. Cohen and M. J. Grossman. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 92–94.)—The use of mixtures of solid carbon dioxide with organic liquids was investigated. Solid carbon dioxide was added to the liquids contained in vacuum flasks until no further reduction in temperature occurred. The following liquids were considered the most suitable for practical use, as they neither crystallised nor became viscous:

| Liquid | Temperature produced °C. |
|-------------------------------|--------------------------|
| Ethylene glycol | –15 |
| Dimethoxytetraethylene glycol | –31 |
| Diethyl carbitol* | –52 |
| Carbitol acetate* | –67 |
| Cellosolve* | –73 |
| Cellosolve acetate* | |
| Diacetone alcohol | |
| Butyl cellosolve* | |

When the lowest temperature was required it was found convenient to use a series of low-temperature baths rather than to place the object at air temperature into the coldest mixture, which might result in violent evolution of carbon dioxide. The same fluid may be used repeatedly, more solid carbon dioxide being added as required. S. G. C.

* Trade names.

Reviews

PHYSICO-CHEMICAL METHODS. Vol. I. MEASUREMENT AND MANIPULATIONS. Pp. xv + 686. Vol. II. PRACTICAL MEASUREMENTS. Pp. ix + 580. By JOSEPH REILLY, M.A., D.Sc., F.Inst.P., F.I.C., and WILLIAM NORMAN RAE, M.A., Sc.D., F.I.C. London: Methuen & Co., Third Edition. 1940. Price £4 4s.

All chemists use physical methods to some extent, and such instruments as the polarimeter and microscope are to be found in every laboratory. The spectrograph is taking a prominent place in the larger laboratories, and physico-chemical methods are being utilised more and more, not only for research but also for routine analytical purposes. Some physical instruments may have a relatively small application outside certain specialised fields; but others, *e.g.* the polarograph, have a very much wider scope than might be guessed by those unacquainted with them. It is often complained that physical apparatus is very expensive. This may be true for complete, ready-to-use instruments, housed in polished cases; but when the physico-chemical principles of a method are understood, a "hook-up" will serve, nay, may be preferable, since it can be modified to suit particular conditions. Many physico-chemical processes need no special apparatus other than that which any worker can make for himself.

The practising chemist needs to be able to keep abreast of the progress in allied sciences, to refresh his memory on the principles underlying the methods used, and to consider their adoption for his particular purposes. The work under review is an admirable source for satisfying these needs in respect of physics. Volume I covers the general plan of a laboratory; measuring units; statistical and graphical representation of results; apparatus; manipulative operations; high pressure technique and some particular physical measurements. Volume II includes separations involving distillation, crystallisation, centrifugal and other mechanical processes; optical and electro-chemical measurements; indicators and *pH* measurement; applications of the thermionic vacuum tube; dielectrics and radioactivity. These are only the main headings, for the subject matter is, as the preface says, "an orderly collection of borderland problems wherein both physicists and chemists co-operate." Each volume contains over 400 diagrams.

Again to quote the preface, "the temptation to convert the work into an unwieldy encyclopaedia has been resisted." For this the authors are to be congratulated, since it has not been coupled with a reduction of the number of methods dealt with, but only with the number of pages devoted to each. Some parts may be found too concise for practical purposes; but the facts are there, and many references are given to suitable books and original papers for further reading on the subject matter of each chapter.

In each volume the index occupies parts of two pages only. This has been done deliberately to avoid making it, as a note says, "so detailed as to be clumsy and ineffective." In the reviewer's opinion it is now ineffective because it is so lacking in detail, being of little more value than the table of contents. The

arrangement of the subjects is such, however, that the interested reader will soon find his way about without an index.

No modern chemical laboratory can afford to be without these two volumes. Although the price is high, it must be equated to the comprehensive nature of the subject matter.

J. R. NICHOLLS

BELL'S SALE OF FOOD AND DRUGS. Tenth Edition. By R. A. ROBINSON, O.B.E., Barrister-at-Law, Chief Officer, Public Control Department, Middlesex County Council, and ROBERT IVES, Barrister-at-Law. Pp. xxvii + 363. London: Butterworth & Co. (Publishers), Ltd., and Shaw & Sons, Ltd. 1939. Price 15s. net.

"This little volume is intended to supply a want which it is believed has been felt by persons who are engaged in carrying out the Sale of Food and Drugs Acts. Great care has been taken to include all cases bearing upon the working of the Acts, and also those in which disputed questions of a chemical nature have arisen. It is hoped that the book may be found useful not only to those upon whom the working of the Act devolves, but also to the trade and general public." These words written in the preface to the first edition of "Bell's Sale of Food and Drugs" seem to have been almost prophetic, and the hope expressed so long ago as 1886 has been more than well fulfilled.

The issue of a new edition, the tenth, has been made necessary by the enactment of the Food and Drugs Act, 1938, which came into force on October 1st last. This edition has had to be considerably enlarged, as the new Act includes sections relating to a number of subjects which were previously to be found in the Public Health Acts, such as unsound food, food poisoning, the sale of horse-flesh and the cleansing of shell fish. Even with this enlargement, it has been possible still to retain such statutes as the Merchandise Marks Act, 1926, and the Sale of Food (Weights and Measures) Act, 1926, as in the eighth and ninth editions, and to include the Agricultural Produce (Grading and Marking) Acts, 1928 and 1931.

Mr. Robinson, who has been responsible for the preparation of five earlier editions, has now introduced Mr. Ives, also a barrister-at-law, as a co-editor of this authoritative work. The bulk of the present edition comprises the Food and Drugs Act, 1938, carefully annotated, together with the text of the other Acts cited above. A table of replacements shows what provisions have been repealed, where they may be found in *Halsbury's Statutes*, and the corresponding sections of the previous Food and Drugs Acts. Part II deals with Departmental Regulations and Orders.

In this edition 42 cases previously reported in the ninth edition have been omitted and 38 fresh cases inserted. The chemical notes have been augmented with new data, including such references as a suggested maximum of 40 per cent. of water in processed cheese and the inclusion of cooking fats with lard. The possible presence of ailanthus in mint is mentioned, and under the heading Port and Madeira wine there are references to two High Court cases—*Holmes v. Pipers Ltd.* (1914) and *Sandeman v. Gold* (1924). A paragraph on whisky is also included.

Regarding meat, apparently an experienced inspector is expected to identify different kinds of liver simply by making a cursory glance without reference to the

Public Analyst. Mr. Robinson's views on a possible waste of public money on analyses may be observed on page 5, and no doubt the Public Analyst will await with interest the case in which the vendor, having admitted a sale of margarine as butter, declares in Court that it was butter after all. Perhaps the Public Analyst would be permitted to smile, as he certainly might do, at Mr. Robinson's belated discovery that methods other than chemical may be employed.

Little criticism is needed for a book that has reached its tenth edition, and it is pleasant to note that such mistakes as were made in the ninth edition appear now to be corrected. It is safe to say that both Public Analysts and lawyers engaged in work dealing with Food and Drugs certainly owe a debt of gratitude to the two co-editors for bringing this invaluable book up to date.

The publishers propose to issue, from time to time, supplements to this work, and a pocket inside the back cover is provided to hold them.

ERIC VOELCKER

A HISTORY OF CHEMISTRY. By F. J. MOORE, Ph.D. Revised by William T. HALL. International Chemical Series. JAMES F. NORRIS, Ph.D., Consulting Editor. Third Edition. Pp. 429. Index, pp. 47. London: McGraw-Hill Publishing Co., Ltd. 1939. Price 20s.

In common with similar histories this volume devotes its early pages to a description of the events preceding the birth of chemistry as a specific entity. The science of chemistry evolved gradually from the practice of art as applied to metallurgy; its progenitors in the investigation of natural substances included philosophy, astrology, necromancy, religion and chicanery, and its main aims were the search for the elixir of life and the philosopher's stone—tempting baits for men who sought wealth and notoriety. Such chemical knowledge as there was up to the fifteenth century was termed alchemy.

The author has dealt with this part of the subject lucidly and in interesting chapters which lead to the *renaissance* period, when Paracelsus, Agricola, Valerius Cordus, Basil Valentine, Van Helmont and Johann Glauber appear in a description of their work and influence on the subject before Boyle, Mayow and Stahl became prominent and the Phlogiston Theory held the stage for so many years.

This period was the immediate forerunner of the anti-phlogiston era, when Black, Cavendish, Scheele and Priestley made their all-important discoveries, and the recognition of oxygen as an element may be considered to mark the time when more modern ideas were needed to fulfil all the precepts which led us to the fundamental principles of eighteenth-century chemistry.

From this period the author takes his readers consecutively through the work done by Lavoisier and Berthollet to the time of Dalton and the Atomic Theory, and here he shows how the physical properties of matter became an integral part of the study of chemistry. Galvani, Volta and Humphrey Davy made such alterations in the existing ideas and converted them into conceptions so novel that one is hardly surprised at the developments which followed. Organic Chemistry, and all that it foreshadowed, was a new sphere of chemistry from which, during the past century, the whole of mankind has benefitted.

The chapter dealing with Organic Chemistry since 1860 is full of interest. The scientists' names are too numerous to quote, but every chemist is familiar with them in connection with reactions or processes or principles. Many of them are living no longer, but it is of interest to read of the modern work of Haworth on sugars, that of Hans Fischer on physiological chemistry, and that of Karrer on the vitamins. The final chapter in the book is a lengthy one of a hundred pages, so full of the progress of Inorganic Chemistry since 1860 that it deserves a volume apart.

The volume is fully illustrated, and very many of the portraits will be familiar to most English readers. Such references and dates as could be checked are correct, and the volume should be indispensable to students of the subject; it is full of profoundly interesting material for the modern chemist, be he specialist in the organic, inorganic or physical department of the science. C. EDWARD SAGE

AN INTRODUCTION TO BIOCHEMISTRY. By Prof. W. R. FEARON, M.A., Sc.D., M.B., F.I.C. Second Edition. Pp. xii + 475. London: William Heinemann (Medical Books), Ltd. 1940. Price 17s. 6d. net.

It is almost exactly six years since the first edition of Professor Fearon's gallant book was reviewed in this Journal (*ANALYST*, 1934, 59, 372). Much biochemical water has flowed under the bridges of science since that time—and Professor Fearon has faced with renewed gallantry the incorporation in his second edition of all relevant new knowledge gained in the intervening period, though this involved rewriting three-quarters of the book and increasing its length by more than one-half. The price has increased about proportionately—less than might have been expected in present circumstances.

There are very many text-books of biochemistry written in English (more or less), but curiously few written and published in Great Britain and Eire. It is not an exaggeration to say that this one, with the additions, improvements and corrections made since the first edition, is now the best of the home products and a dangerous competitor to the best of the foreign—Bodansky's "Introduction to Physiological Chemistry" (see *ANALYST*, 1938, 63, 921).

A work of this kind, having been brought as nearly up-to-date as exigencies of publication permit, is unlikely to be free from undetected slips of phraseology and expression, or to have avoided minor inaccuracies in a few places; still less is it likely that every statement of opinion in it will find general acceptance among biochemists. Indeed, if it did, it is probable that its author might feel that he had in some manner betrayed his Irish nationality. A review is no place for calling attention to these minutiae, except in a general way; individually they are better brought to the author's attention by personal correspondence, especially to an author like Dr. Fearon, who has wisely shown himself unusually willing to accept and act on the friendly criticisms of his many favourable reviewers.

It is, however, necessary for a reviewer to show that he has done something more than "break the back of the book"—which Mr. Belloc's "Caliban" advised as the only essential part of book reviewing. I note that Dr. Fearon uses D- and L- to denote the structural configurations of compounds in relation to their positively and negatively rotating chemical ancestors, reserving the conventional

d- and *l*- to express actual rotation. It is a little difficult to see what advantage this has over the use of *d*- and *l*- for structure and of (+) and (−) for rotation. Moreover, when it comes, for example, to the amino-acids, Dr. Fearon avoids the problem altogether by not crediting them with any optical activity at all, although he has a general statement on the subject early in Chapter IX. What the student needs to know is *both* the configuration of the amino-acids (where this has been established) *and* the directions of their optical rotations. I would suggest that, when revising this book, the author should include in his discussion of fat absorption some consideration of the work of Frazer and Stewart; the implications of this are so disturbing to current complacency about complete intestinal lipolysis that they deserve study for that reason alone—apart from the fact that Frazer's views on the subject are almost certainly correct. One other comment—also on a point of detail. Surely Dr. Fearon is wrong in attributing to Karrer (by implication) the view that “condensation of two phytol residues would in theory yield lycopene” (p. 204). The equation for this reaction would be



But the formula for lycopene is $C_{40}H_{56}(13F)$; the condensed molecule needs 11 dehydrogenations before it can have a carotenoid structure.

Leaving lipids, I can find points of dissent about water-soluble substances, also. The most usual biological test for vitamin B₁ (aneurin) is by rat-growth (weight increase), and is the only practical one *not* mentioned by Dr. Fearon. Again, the author appears completely to accept Kögl's claims to have found optically abnormal isomers among the amino-acids of tumour tissues, although the matter is being hotly contested by experts in protein chemistry, and must at the least be held still to be *sub judice*.

The book contains a certain number of directions for actual chemical tests, doubtless for convenience where it is used as a class text-book, but their number is relatively small and it might be better either to omit them altogether—for the general biochemical user they are an embarrassment rather than a convenience—or to relegate them to an appendix.

I know of no book more suitable to the analyst—or any non-biochemist—who wishes to revive his knowledge (or create it) of modern biochemistry, and of none more handy for the biochemist as a working book of reference, in which respect it sets a very high standard for completeness, lucidity and accuracy.

A. L. BACHARACH

FOOD VALUES AT A GLANCE. By V. G. PLIMMER. Pp. 190. London: Longmans, Green & Co. 1939. Price 6s. net.

Anyone who has ever tried to set out the elements of nutrition in a way sufficiently simple to be understood by non-scientific readers knows the difficulty of the task. The degree to which Mrs. Plimmer has succeeded is proved by the publication of this second edition of her short book. The essential feature of the volume, from which it derives its title, is a series of 26 coloured charts which show graphically the composition of 175 common foodstuffs with respect to protein, fat, carbohydrate, mineral salts, moisture and physiologically unavailable material,

and also indicate the proportion of vitamins A, D, B₁, B₂ and C. It is unquestionable that to these clear and brightly coloured diagrams the book owes a large measure of its popularity. Besides these there are a number of black and white diagrams illustrating such aspects of dietetics as the calorie requirement at different ages, the comparative protein value of food in shillingsworths (with, alas, corned beef well in the lead) and many others. Six charts, new to this edition, compare the amounts of vitamins in different foods.

In the text, the chapters dealing with the elements of nutrition are also clear and easily understood. The rôle of each food constituent is described in turn, with the amount of each needed in the diet. The typical daily bill of fare, for a family or school or institution, comprising these nutritional factors is then worked out. All this is excellent. But a text-book, however small and however "popular," must jealously guard against the intrusion of opinion or the most innocent of generous enthusiasms into a realm where facts alone should rule. Perhaps, for example, appendicitis, colitis and cancer may be due to other causes than "the consumption over many years of vitamin-poor foodstuffs."

While the greater part of the book is derived from the first edition brought up to date, certain additions have been made. The inclusion of an index can be given unreserved welcome. Three additional chapters at the end of the volume, however, represent in its most acute form that slide from the scientific world into the political arena which has already been deprecated in a work of this nature. We can sympathise with Mrs. Plimmer for disliking the agricultural policy of the Government; for disapproving of the business principles under which food is distributed in a capitalist England; for condemning the tipping of town sewage into the sea; but do these views enhance the value of an elementary primer on food values?

From the point of view of the analyst the book has many useful features. The charts briefly summarise the composition of foods and can be a quick aid to memory. The tables of daily human requirements, ranging from calories to vitamins, are also valuable for rapid reference. Though it is strange to see pork demonstrated in the chart as containing no vitamin B₁, while ham, bacon and pork-pie possess it, inaccuracies appear to be few.

"There is probably no field of human thought in which sentiment and prejudice take the place of sound judgment and logical thinking so completely as in dietetics," quotes the author. As they put down the book certain analysts of our acquaintance may sigh. Alcoholic drinks are a common cause of obesity; continental cooking may lead to avitaminoses; we know that tinned salmon, brown bread and cabbage would make a perfect diet—but, oh, how we wish they would not.

MAGNUS PYKE

MANUEL DU SAVONNIER. By A. MATAGRIN. Pp. xvii + 268. Paris: Gauthier-Villars. 1938. Price 30 fr.

This book contains no preface, and there is no indication of the class of reader for whom it is intended. It opens with a very interesting chapter dealing with the early history of soap-making, and with the economic position of the industry in the principal countries of Europe, but the book, as a whole, rather gives the impression

of being written by an author without any very intimate knowledge either of soap-making or of its chemistry.

So far as the description of actual soap manufacture is concerned, the book follows conventional lines and deals, rather superficially, with the preparation of most types of soap, including hard, soft and liquid, household, textile, toilet, and medicated soaps. It may be remarked that 20 hours for the pasting stage (p. 63) appears an extraordinarily long time to allow for this operation. Modern developments, such as attempts to make the saponification process continuous and the use of ethanolamines and of antioxidants, are included. The plant used in the various operations is fairly fully illustrated, but, although the figures are mainly of machinery produced by one of the best-known French soap engineers, they are rather poorly reproduced.

The treatment of the theory of detergency is most inadequate, and the book concludes with a section, of seven pages, on the analysis of oils and soaps, which is so unreliable as to be worse than useless. Thus, the strengths of semi- and decinormal solutions of potassium hydroxide are given as 112 g. per litre and 56 per cent. respectively, the acid value is said to be the Koettstorfer value, and the unsaponifiable matter in a fat is to be obtained by determining the fatty acids after saponification and subtracting the percentage of these from 100. In the analysis of soap no distinction is made between unsaponified fat and unsaponifiable matter, nor between caustic alkali or other free alkali, the free alkali being obtained by salting out the soap, filtering, and titrating the alkali in the filtrate.

W. H. SIMMONS

Publications Received

KINGZETT'S CHEMICAL ENCYCLOPAEDIA. 6th Ed. Revised and Edited by R. K. STRONG. With Foreword by Sir GILBERT MORGAN. Pp. x + 1088. London: Baillière, Tindall & Cox. 1940. Price 45s.

CHEMICALS OF COMMERCE. By E. D. and C. T. SNELL. Pp. viii + 542. London: Chapman & Hall. 1940. Price 28s. net.

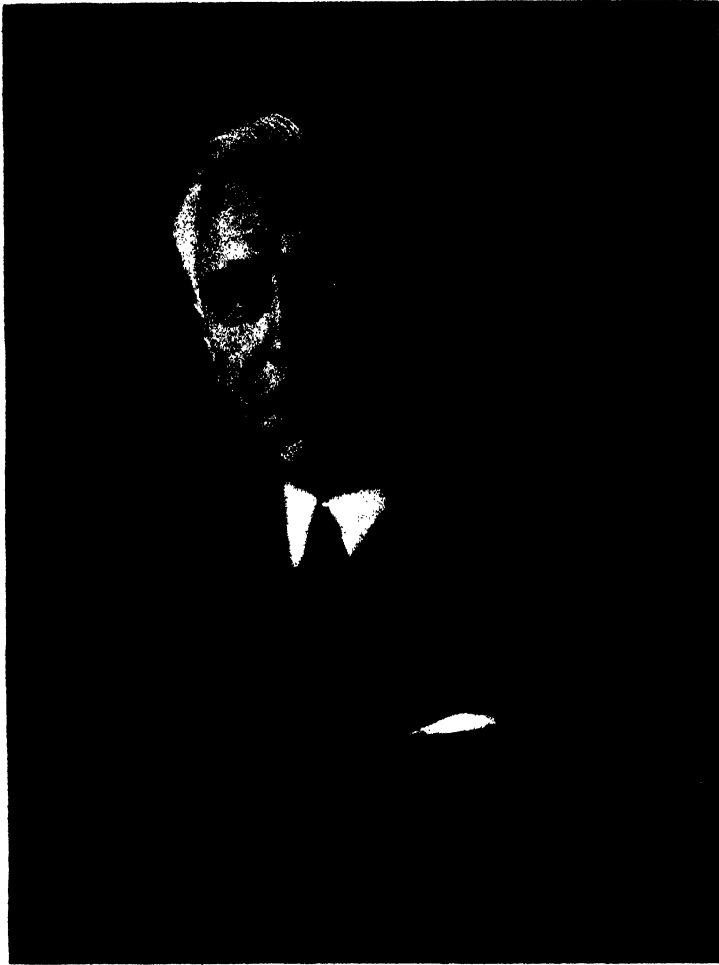
COLLOID CHEMISTRY. By H. B. WEISER. Pp. viii + 428. London: Chapman & Hall. 1939. Price 24s. net.

LES PARFUMS NATURELS. By Y. R. NAVES and G. MAZUYER. Pp. xvi + 398. Paris: Gauthier-Villars. 1939. Price 120 fr.

POISONS: THEIR ISOLATION AND IDENTIFICATION. By F. BAMFORD. Pp. viii + 344. London: Churchill. 1940. Price 18s.

A TEXT BOOK ON LIGHT. By A. W. BARTON. Pp. x + 426. London: Longmans, Green & Co., Ltd. 1939. Price 8s.

ANALISI DEI METALLI NON FERROSI. By I. COMPAGNO. Pp. xii + 493. Milan: Ulrico Hoepli. 1939. Price Lire 60.



Subjects Ltd photographers

Emerging/Whitney Ltd pho

W.H. Roberts

PRESIDENT 1938-1939

THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

Obituary

FRANK GEORGE EDMED

FRANK GEORGE EDMED died at Southsea on January 22nd, at the age of 63, only three weeks after his retirement from the post of Admiralty Chemist, which he had held since 1926, when he succeeded Mr. Arnold Philip. Edmed was born at Brighton and educated at Brighton Grammar School. Later he was a student at the Royal College of Science, obtaining the Associateship in 1897, in which year he also graduated B.Sc. (Lond.) with honours in chemistry. After a short service on the research staff at the Jenner Institute, Edmed joined the staff of the War Department Chemist at Woolwich; later (1919) he became Superintending Chemist in the Inspection Department of the Royal Naval Cordite Factory, and continued in this post until his appointment as Admiralty Chemist.

Edmed's experience in analytical chemistry, gained during his many years' service under the War Office and the Admiralty, was very extensive; few chemists, indeed, could claim such wide experience. He served on numerous technical committees of the British Standards Institution and the Institute of Petroleum, and his wide knowledge and experience was of the greatest value. He was particularly devoted to the work of the Institute of Chemistry. He served on the Council for three periods: 1928-31; 1932-33 and 1937 to the time of his death, and from 1933-36 he was a Vice-President. In addition, he was for some years Chairman of the London and South-Eastern Counties Section. He was on the Council of this Society in 1935 and 1936.

In recognition of his services during the Great War he was appointed an Officer of the Most Excellent Order of the British Empire.

Edmed had a great sense of humour and a gift of ready repartee. His cheerful outlook on life endeared him to his many friends and associates. He leaves a widow and two daughters.

J. S. S. BRAME

THOMAS STANTIAL GLADDING

THOMAS STANTIAL GLADDING was born in Virginia in 1853 and, while he was still a child, his family moved to Providence, Rhode Island, so that his background was New England. He was educated in the public schools of Providence and at Brown University, where he graduated B.A. in 1875 and M.A. in 1878. He was a brilliant scholar, stood high in his class, and was elected to the honorary society of Phi Beta Kappa.

He taught chemistry for two years at Suffield Academy and for one year at Worcester Academy. In 1878 he went to New York City and formed with Charles M. Stillwell the partnership of Stillwell and Gladding, a professional firm for chemical consultation and analysis.

Gladding was active in his profession for many years until his retirement in 1932. He was especially able in the development of analytical methods, increasing their accuracy and availability for practical use. The two outstanding methods that he developed and improved were the neutral ammonium citrate method for the determination of the availability of the combined phosphoric acid in fertilisers, and the potassium platonic chloride method for the determination of potash. These methods have been adopted as official by the Association of Official Agricultural Chemists of the United States.

For many years Gladding had been a member of this Society, the American Chemical Society and the Society of Chemical Industry. In addition to his professional work he was active in the church, in the Young Men's Christian Association, and in civic matters in his home town of Montclair, New Jersey. He was fond of travel and travelled extensively before the World War. After his retirement from active work he lived in Easton, Maryland, until his death in December, 1939, in his 87th year.

He leaves a widow but no children.

H. E. CUTTS

FRANK THOMAS SHUTT

FRANK THOMAS SHUTT was born in Stoke Newington, London, England, on September 15th, 1859, a son of Denis Shutt, C.E., and Charlotte Cawthorne Shutt. After receiving private tuition in England he came to Canada as a boy and was a pupil of Dr. William Hodgson Ellis at the School of Technology, Toronto. On removal of this school to Queen's Park to become the School of Practical Science of the University of Toronto, Dr. Ellis became Professor of Applied Chemistry and acted as Public Analyst. As his assistant for five years Shutt obtained valuable training and experience in analytical chemistry.

In 1885 he obtained his B.A. from the University of Toronto with first-class honours in chemistry, and he received his M.A. one year later. In 1914 he was awarded the D.Sc. (*honoris causa*). On the formation of the Dominion Experimental Farms System, with headquarters at Ottawa, he was chosen to be the first chemist in 1887. In 1912 he received the title of Dominion Chemist and was also appointed Assistant Director. He served in this dual capacity until his retirement in 1933.

During his forty-six years of service Shutt made many valuable contributions to agriculture. Always interested in the practical side of farming, his guiding thought was the application of chemistry to the everyday problems of the farmer. He was a pioneer in Canada in establishing on a firm scientific basis the manurial value of clover and legumes. His investigations with fertilisers have gone far to bring about their rational and judicious use. In 1901 he published results of an extensive study of the "soft pork" problem, which has proved a valuable guide to all who are interested in pork and bacon production. His investigations of the qualities of Canadian-grown wheats were extensive and had much to do with the permanent establishment of Marquis wheat. His publications in scientific journals were numerous, and he frequently contributed articles of practical value to the agricultural press.

Dr. Shutt's achievements in scientific agriculture have been widely recognised. In 1935 he received the honour of the C.B.E. The Royal Society of Canada awarded him the Sir Joseph Flavelle medal, and he received the prize of the American Society of Agronomy for outstanding research. The Canadian Institute of Chemistry recognised his services by electing him to an honorary fellowship. He was a Fellow of the Chemical Society, the Royal Society of Canada, and the Institute of Chemistry of Great Britain and Ireland. He was elected to our Society in 1916, and he was also a member of the Society of Chemical Industry, the American

Chemical Society and the American Association for the Advancement of Science, and an original member of the Association of Official Agricultural Chemists.

He passed peacefully to his rest at Ottawa on January 5, 1940, in his eighty-first year. His contributions to agriculture in Canada remain a lasting memorial.

Apart from his profession Dr. Shutt's main attachment was to his family. He remained a bachelor, and was devoted to his mother, a charming, stately old lady, who for many years was the head of his home. On her death his sister took her place. For her and his brothers, his nephews and nieces, "Uncle Frank" was the centre and chief tie of union of the family.

Next to his family, probably, the deepest current of his life set to his church. He was never a man who talked about his religious convictions, which by training and habit formed as natural and integral a part of his life as breathing. He had a great many acquaintances and a small circle of warm friends. Talks over a pipe before a grate fire, walks in the country, photographing expeditions, and little picnic parties with tea made over an outdoor fire were some of the ways in which he enjoyed comradeship.

His life was enriched by a number of hobbies. He was fond of music; indeed, for several years he played the organ in an Ottawa church. Latterly, he derived much pleasure from the gramophone and the radio. He was to the end an ardent photographer, and some of his work attained a high degree of excellence.

His was a thoroughly conservative nature. In politics he took no part, but in music, in art, in thought and in manners he held to the old ways. Fastidious in his habits, in his speech, and his writings, he would, we think, have liked the designation which might well be applied to him, "A gentleman of the old school."

C. H. ROBINSON

W. J. SYKES

Hair Dyes II. The Functions and Reactions of Phenols

BY H. E. COX, D.Sc., Ph.D., F.I.C.

(Read and illustrated by a cine-film in colour at the Meeting, April 3rd, 1940)

It is a commonplace to hear it said that a particular hair dye contains a certain diamine, just as though that were the whole of the story. Such is very far from the truth; tinctorial power is obviously important, but there are many other desiderata. For example, an ideal hair dye should (1) provide a silky lustre on the hair; (2) not make the hair brittle; (3) not irritate or stain the scalp; (4) not fade or produce "off" colours; (5) be unaffected by permanent waving or the application of heat or alkali. Moreover, it will be required to produce a wide range of colours to suit the particular individual. Such range of colours cannot be obtained by oxidation of any one diamine, aminophenol or other compound. It is probably true to say that the perfect hair dye which fulfils all these conditions does not yet exist. Those who have studied the subject and examined a number of types of hair dyes will know that a good hair dye of the oxidation class will contain, in addition to the diamine, buffer salts, wetting agents—soap or otherwise—perfume, perhaps alcohol or glycerin, and, most important, a quantity of polyhydric phenol such as resorcinol, catechol or pyrogallol. The range of colours is usually attained by the adjustment of the relative quantities of the base and the phenol. So far as I know, the functions and reactions of the phenol have not been examined, although the practical dyer or the manufacturer would say he could not get his shades without them.

There are certain factors which complicate the study of hair dyes. Hair has the general properties of wool so far as composition is concerned, but it has considerably less affinity for dyes. Living hair, too, is more resistant than non-living hair; it is apt to be protected by natural grease, not removed by shampooing, which resists the absorption of dyes just as do living yeast cells. So, while the use of wool or of dead hair is very useful for studying the reactions and general colours produced, precise results can only be ascertained by trial on living hair. In general, hair dyes have to be oxidised at temperatures below, say, 37° C., usually in weak alkaline solutions by the application of diluted hydrogen peroxide or solutions of urea peroxide. The oxidation must be neither too fast nor too slow. It is clear that direct oxidation methods involving the use of heat or of dichromate or ferric chloride will not be appropriate to the study of the chemical reactions; such reagents produce green or blue indamines or indophenols in presence of amines or phenols and would be most unwelcome on a lady's head.

It will be convenient to consider primarily the oxidation products of *p*-phenylenediamine under various conditions, since this substance, notwithstanding certain well-known disadvantages, is still the most efficient hair dye and is capable of producing the most natural shades. Excess of a solution containing about 2 per cent. will produce a black, and less quantity will give intermediate shades of grey when oxidised with hydrogen peroxide.

EXPERIMENTAL

Although the shades obtainable with living hair differ in certain respects from those obtainable with wool, it is convenient to use wool for experimental purposes, and it seems that the reactions are the same in kind. Preliminary experiments show that the colour obtainable from a given mixture depends upon three factors: the particular base and phenol present, the relative quantities of these substances, and the time of application. The rate of oxidation is comparatively slow at hair-dyeing temperatures; so, too, is the rate of absorption; it is slower with live hair than with dead hair, but more rapid with wool, and the normal duration of application of a hair dye is to be reckoned in minutes. It was found, for example, that 1 per cent. *p*-phenylenediamine dyed wool a blue-black colour in 15 minutes, the intensity of the colour being proportional to the time, but the result is always grey or black. When a half-molecular proportion of resorcinol is added the result in the same time is a brownish-black; when an equal molecular proportion of resorcinol is used the product is a rich brown and is not changed by further addition of resorcinol. This indicates that one molecule of the amine and phenol are jointly concerned in the reactions described later. In order to trace the course of the reactions more accurately and determine how much of the reactants was absorbed, each of the following experiments were made at room temperature on 10 g. of white wool; the mixtures were allowed to act for 24 hours, so that all reactions might be complete and colours fully developed.

A. *p*-Phenylenediamine only.—Dye bath:—*p*-phenylenediamine, 1.08 g.; sodium carbonate (dry), 1.0 g.; water, 150 ml.; hydrogen peroxide (20 vol.), 20 ml.

The dyed wool had a dull black colour, and the diamine had distributed itself thus:—*p*-phenylenediamine combined in the wool, 0.405; as insoluble (Bandrowski) base, 0.365; unoxidised remaining in the solution, 0.311; total, 1.081 g.

Examination of the black dyed wool showed that it contained free Bandrowski base (extractable by pyridine) and an azine combined in the fibre. Only this latter is the real dye; the whole of the Bandrowski base can be extracted, leaving an uninteresting dull black.

B. *p*-Phenylenediamine and catechol.—Dye bath:—*p*-phenylenediamine, 1.08 g.; sodium carbonate, 1.0 g.; catechol, 2.20 g.; water, 150 ml.; hydrogen peroxide, 20 ml.

Wool was dyed a dark brown colour and the distribution was:—*p*-phenylenediamine combined in the wool, 0.353 g.; as insoluble base, nil; unoxidised in the liquor, 0.727 g.

No Bandrowski base had been formed; there was no soluble dyestuff, but a brown-black dye of azine-type had been formed.

C. *p*-Phenylenediamine and resorcinol.—Dye bath:—*p*-phenylenediamine, 1.08 g.; sodium carbonate, 1.0 g.; resorcinol, 2.20 g.; water, 150 ml.; hydrogen peroxide, 20 ml.

The wool was dyed a deep brown colour and the base was distributed as follows:—combined in the wool, 0.322 g.; as insoluble base, 0.074 g.; incompletely oxidised in solution, 0.698 g.; total, 1.094 g.

The wool did not contain any Bandrowski base; it yielded nothing to pyridine and the dark brown dye gave the reactions of an azine or oxazine.

D. *p*-Phenylenediamine and quinol.—Dye bath:—*p*-phenylenediamine, 1.08 g.; sodium carbonate, 1.0 g.; quinol, 2.20 g.; water, 250 ml.; hydrogen peroxide, 20 ml.

The wool was dyed a cocoa-brown colour and contained the equivalent of 0.349 g. of diamine. There was no formation of Bandrowski base, but some insoluble matter was formed unless the mixture was well diluted. The explanation is that *p*-phenylenediamine forms a crystalline addition compound (m.p. 199–200° C.) with quinol; this contains 1 mol. of quinol and has low solubility, so that it is necessary to add more water.

E. *p*-Phenylenediamine and pyrogallol.—Dye bath:—*p*-phenylenediamine, 1.08 g.; sodium carbonate, 1.0 g.; pyrogallol, 2.52 g.; water, 150 ml.; hydrogen peroxide, 20 ml.

The wool was dyed a light brown colour; it contained the equivalent of 0.356 g. of diamine. There was no Bandrowski base.

F. *p*-Phenylenediamine and phloroglucinol.—Dye bath:—*p*-phenylenediamine, 1.08 g.; sodium carbonate, 1.0 g.; phloroglucinol, 2.52 g.; water, 150 ml.; hydrogen peroxide, 20 ml.

Wool was dyed a greyish-brown colour and contained the equivalent of 0.247 g. of the diamine. There was no insoluble base.

EXAMINATION OF DYED WOOLS.—The dyed wools were examined to ascertain, as far as possible, the chemical type of dye formed and the distribution of the diamine and phenol. With the *p*-phenylenediamine alone it is not difficult to identify Bandrowski's base and a complex azine in the fibres. With catechol and resorcinol two substances can be detected—an (oxazine) dye and a diamine-phenol addition product, loosely held in the fibres, which can be extracted. The dye can be reduced with formaldehyde sulfoxylate reagent, and quantitative extraction of the residual liquor in the dye bath shows that approximately equal molecular proportions of the phenol and the base have been absorbed and oxidised. The colour can also be dissolved out by conc. sulphuric acid in which it forms the characteristic green solution, becoming brown again on dilution; the solutions may be reduced with zinc dust and re-oxidised in the usual way.

With the trihydric phenols somewhat different reactions appear to take place. There is a marked difference in colour, as light browns are produced, although the fibre has absorbed approximately the same amount of diamine. This brown colour, however, can be further oxidised to dark brown and black by means of dichromate applied to the fibres; thus it seems that the trihydric phenols retard the oxidation of the diamine and form, in addition to the dyestuffs, some rather unstable quinones or hydroxyquinones which are capable of further oxidation to black or brown substances.

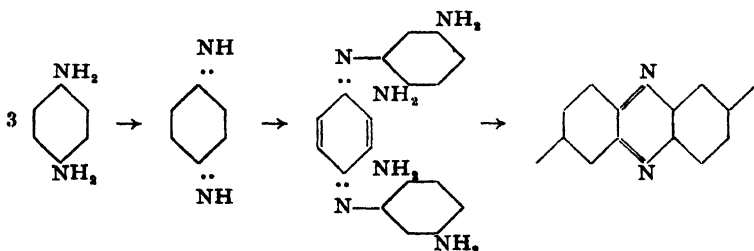
BEHAVIOUR OF OTHER PHENOLIC COMPOUNDS.—In order to observe whether these results are at all peculiar to *p*-phenylenediamine a similar set of experiments was made with *p*-*p'*-diamino-diphenylamine. With resorcinol and catechol similar results were obtained—that is, good dark browns, but with trihydric phenols the

resulting colours were all comparatively light browns and, particularly with pyrogallol, the effect of the phenol was much greater than would appear from its chemical equivalent. Even one-tenth or one-fifth molecular proportion had quite a marked effect. It is evident that both dihydric and trihydric phenols prevent the formation of Bandrowski's base and that resorcinol and catechol facilitate the formation of oxazines or oxazones, which are the desirable and permanent colouring matters. Quinol also leads to the formation of an oxazine, but perhaps by a slightly different route.

The colours can be reduced with Formosul reagent and re-oxidised in air, and no phenol can be separated by processes of extraction, even after reduction.

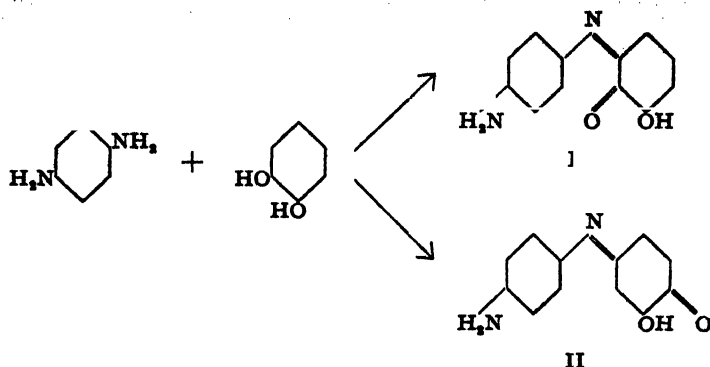
THE REACTIONS INVOLVED

As has been shown in a previous paper,¹ there is little doubt that when *p*-phenylenediamine is oxidised by hydrogen peroxide in alkaline solution in presence of animal fibres, the principal products are Bandrowski's base, an azine and a little quinone. I have shown too that the azine can be produced by direct oxidation of the Bandrowski base. These reactions are commonly represented thus:

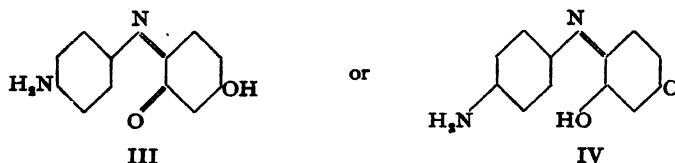


The final product must have a molecular weight of at least 500, and may be reduced to a colourless leuco compound. In presence of catechol, resorcinol and the other phenols there is no formation of insoluble base, so that the reaction must proceed otherwise. This may be due in part to the fact that most of the phenols form double compounds with the diamines; some of these are of commercial importance. For example, *p*-phenylenediamine with catechol forms a white crystalline product containing two mols. of catechol and having m.p. 110° C.; similarly with quinol there is formed a well-defined compound with m.p. 200° C., containing one mol. of each constituent. Such compounds preclude the formation of Bandrowski's base, and the fact that they are stable enough to prevent the direct oxidation of the diamine is shown by the fact that hypochlorite produces no precipitate of dichlorimide, as it does with the diamine alone. Oxidation in this way of mixtures or of the double compound gives rise to red and violet dyes, which are not very stable but are rapidly oxidised or condensed to brown substances. It is commonly stated in textbooks that indophenols are blue, but all are not so; Hodgson and his collaborators² have described several red and brown ortho-indophenols.

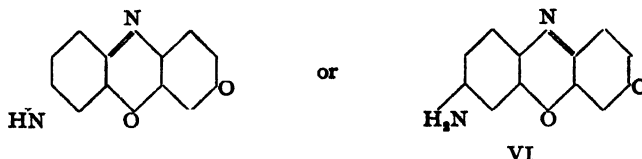
Further, the indophenols are unstable compounds which are easily oxidised or decomposed in various ways, although some of the ortho-compounds may even be acetylated in presence of a reducing agent. The essential factor determining the colour of an indophenol is that the ortho-compounds are red or brown, whereas the para-compounds are blue.³ As these colours are observed in the course of the oxidation of mixtures, it appears that the reactions proceed *via* the indophenols. Thus, when a catechol is oxidised with a *p*-diamine or condensed with *p*-nitrosodimethylaniline there are two possibilities:



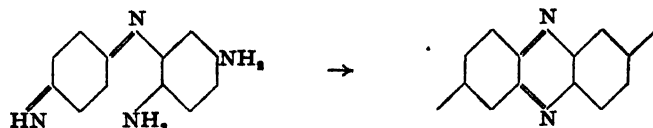
Of these, I will be brown and II blue; similar results may be expected from resorcinol. III will be red, and IV will be blue.



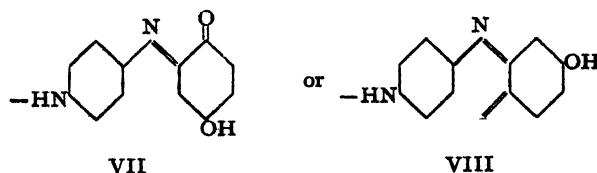
If these red and brown indophenols remained unchanged, the problem would be fairly simple; but, although they are more stable than the blue para-compounds, they slowly change to insoluble brown and black substances of high molecular weight, which evidently must be formed by condensation of the ortho-quinonoid compounds, such as I and III. This may be accomplished by the elimination of hydrogen and the formation of oxazones; thus with resorcinol there will be formed



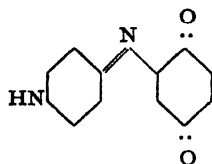
and the molecule may be prolonged. Such a compound would be similar to the black produced on direct oxidation of Bandrowski's base, which is also a complex azine



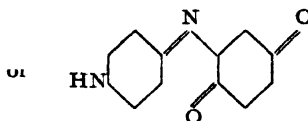
When quinol is used a blue indophenol does not seem possible, as the para-positions are occupied; there are two ways in which a brown compound may be formed:



When the mixture is oxidised a brown compound is formed which quickly changes into an insoluble base. This base, when separated, washed and crystallised from benzene, showed no melting point, but had the ultimate composition $C_{11}H_8N_2O_2$. It is but slightly soluble in water, more so in alcohol, and has the characters of a complex quinone for which two tautomeric formulae are possible:



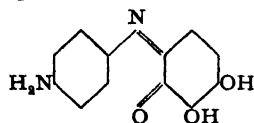
IX



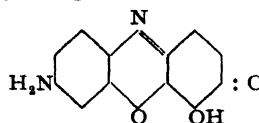
X

and as the final product is an oxazone dye, it seems certain that X is the correct formula.

Evidently such a compound is formed in the fibres of the hair; that this is so is also shown by the fact that hair may be dyed by boiling it in slightly alkaline solution with this brown substance, just as it is when the diamine and phenol are oxidised together on the hair at ordinary temperatures.



XI



XII

Analogous brown compounds, XI and XII, can be formed from pyrogallol and phloroglucinol; for example, pyrogallol can form first a brown indophenol and then an oxazone by loss of one of the hydrogens from the hydroxyl groups. It is evident that there is room for much further study.

CONCLUSIONS.—It is concluded, therefore, that the functions of phenols in hair dye mixtures are:—(1) They promote the formation of fast colours particularly of brown shades. (2) They prevent the formation of Bandrowski's base. (3) They give a better colour for the same amount of diamine. (4) They promote the formation of *o*-indophenols and oxazones in the fibre. (5) The trihydric phenols retard the oxidation of the diamines, form complexes with the base, and promote the formation of light brown colours. (6) These results are brought about mainly by the formation of ortho-quinonoid indophenols which are further oxidised to oxazones.

I hope to describe methods of analysis applicable to some of these mixtures at a later date. In conclusion one debatable point may be mentioned; all the polyhydric phenols are capable of forming addition compounds with *p*-phenylenediamine, usually with one molecular proportion of the phenol. Such compounds have well-defined melting-points, but may be oxidised either in acid or alkaline solution to produce the same results as the oxidation of a simple mixture. Are such compounds salts of *p*-phenylenediamine within the meaning of Schedule 4 of the Poisons Rules, and is the irritant property of *p*-phenylenediamine lessened when it is so combined with a polyphenol?

REFERENCES

1. H. E. Cox, *ANALYST*, 1934, **59**, 3.
2. H. H. Hodgson and D. E. Nicholson, *J. Chem. Soc.*, 1939, 1405.
3. H. Kehrman, *Helv. Chim. Acta*, 1921, **4**, 527.

THE LABORATORY
11, BILLITER SQUARE
LONDON, E.C.3

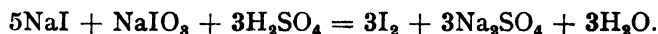
March, 1940

The Iodimetric Determination of Alkali: An Investigation of G. S. Smith's Method

By J. HASLAM, M.Sc., F.I.C., AND R. F. ROBERTS

G. S. SMITH¹ put forward a method for the determination of alkali, in which the alkali solution is heated under reflux with a solution of iodine in benzene for a given period of time, after which the excess of iodine and benzene are removed by boiling, and the solution is cooled. Potassium iodide and sulphuric acid are then added, and the liberated iodine is titrated with standard sodium thiosulphate solution. The method is based on the following reactions:—(i) $2\text{NaOH} + \text{I}_2 = \text{NaI} + \text{NaIO} + \text{H}_2\text{O}$; (ii) $3\text{NaIO} = 2\text{NaI} + \text{NaIO}_3$.

The complete reaction is represented by: $6\text{NaOH} + 3\text{I}_2 = 5\text{NaI} + \text{NaIO}_3 + 3\text{H}_2\text{O}$. On the addition of acid in excess the iodine liberated is equivalent to the amount of alkali which has previously reacted with iodine.



Smith's method is based, to a certain extent, on earlier work of Walker and Gillespie,² who claimed to obtain accurate results by treating the alkaline solution in a conical flask with an excess of *N*/10 iodine solution followed by gentle boiling until all the iodine not required by the alkali had been expelled; the solution was then cooled, and, after addition of dilute acid, titrated with *N*/10 sodium thiosulphate solution.

Smith found that the results obtained by the application of Walker and Gillespie's method were low, and our own experience confirms this. In one actual experiment 25 ml. of *N*/10 sodium hydroxide solution were boiled gently with 50 ml. of *N*/10 iodine solution until the excess iodine was removed; the solution was cooled and treated with acid, and the liberated iodine was titrated with *N*/10 sodium thiosulphate solution. A titration figure of 24.7 ml. of *N*/10 sodium thiosulphate solution was obtained.

This experiment was modified as follows:—Twenty-five ml. of *N*/10 sodium hydroxide solution were boiled with 50 ml. of *N*/10 iodine solution and 1 g. of solid iodine in an open conical flask of about 350-ml. capacity until almost the whole of the iodine had been volatilised, and further additions of iodine in 0.5-g. portions were made until the period of boiling had reached 30 minutes, after which no further iodine was added and the boiling was continued until the excess of iodine was removed. After cooling and treatment with excess of acid, a titration figure of 25.0 ml. of *N*/10 sodium thiosulphate solution was obtained. Although this modification gave an accurate result, it was realised that it would be of little practical use.

Attention should be drawn to the fact that in our application of Smith's method the alkali solutions used in the work have, in effect, been standardised against pure silver,³ as is our usual custom.

Smith's method was applied to a known volume of standard sodium hydroxide solution as follows:—Twenty-five ml. of *N*/10 sodium hydroxide solution, 2 to 3 ml. of pure benzene, a definite weight of sublimed iodine and a known volume of water were placed in a flask of 150 to 250 ml. capacity, and the solution was heated under reflux for a definite time. The excess of iodine was then removed by continuing the boiling after removal of the water from the condenser. The contents of the flask were cooled, and the iodate was determined in the usual manner by means of *N*/10 sodium thiosulphate solution after addition of 2 g. of potassium iodide and 50 ml. of 3*M* sulphuric acid solution.

The results thus obtained under different conditions are given in Table I.

TABLE I

| Expt. No. | Alkali | Volume of solution refluxed ml. | Iodine used g. | Duration of re-fluxing, minutes | Thio-sulphate solution (N/10) required ml. | Notes |
|-----------|---|---------------------------------|----------------|---------------------------------|--|---|
| 1 | 25.0 ml. N/10 NaOH | 70 | 0.5 | 30 | 24.0 | |
| 2 | 25.0 ml. N/10 NaOH | 70 | 0.5 | 30 | 24.0 | |
| 3 | 25.0 ml. N/10 NaOH | 120-180 | 0.5 | 30 | 24.0 | Benzene fractionated previous to use and fraction boiling at 80.5° C. used. |
| 4 | 25.0 ml. N/10 NaOH | 120-180 | 0.5 | 30 | 22.95 | NaOH solution used was free from carbonate and contained a small proportion of barium chloride. |
| 5 | 25.0 ml. N/10 NaOH | 120-180 | 0.5 | 60 | 23.6 | |
| 6 | 25.0 ml. N/10 NaOH | 120-180 | 0.5 | 60 | 23.9 | NaOH solution used was free from carbonate and contained a small proportion of barium chloride. |
| 7 | 25.0 ml. N/10 NaOH | 120-180 | 0.5 | 120 | 23.6 | |
| 8 | 25.0 ml. N/10 NaOH | 120-180 | 0.5 | 180 | 23.3 | |
| 9 | 25.0 ml. N/10 NaOH | 120-180 | 1.0 | 30 | 23.55 | |
| 10 | 25.0 ml. N/10 NaOH | 120-180 | 2.0 | 30 | 23.4 | |
| 11 | 25.0 ml. N/10 NaOH | 120-180 | 0.5 | 30 | 23.8 | Iodine and benzene were added to the boiling solution of the alkali. |
| 12 | 25.0 ml. N/10 Na ₂ CO ₃ | 70 | 0.5 | 150 | 23.45 | |
| 13 | 25.0 ml. N/10 Na ₂ CO ₃ | 70 | 0.5 | 150 | 23.2 | |

It will be noted that the results are low in every instance, and it may be said that in our experience Smith's method has invariably given low results, either at the dilutions which he prescribes or at greater dilutions which we have used to facilitate manipulation. It therefore seemed possible either (1) that the iodine had reacted incompletely with the alkali in the test, or (2) that complete reaction of the iodine with the alkali had taken place, but that a certain amount of reduction of the resulting iodate had occurred, possibly owing to the presence of the benzene; in other words, that the iodate and iodide were not present in the reaction product strictly in the proportion $1\text{NaIO}_3 : 5\text{NaI}$.

The following experiments indicated that the second explanation was correct. Twenty-five ml. of N/10 sodium hydroxide solution, 0.5 g. of iodine, 2-3 ml. of benzene, and about 100 ml. of water were heated under reflux for 30 minutes, after which the excess of iodine was removed by boiling (after removal of the water from the condenser). After cooling, 2 g. of potassium iodide and 50 ml. of sulphuric acid solution were added, and the liberated iodine was titrated with N/10 sodium thiosulphate solution; 23.65 ml. of N/10 sodium thiosulphate were required.

In a duplicate experiment under the same conditions the liquid, after removal of the excess iodine by boiling, was transferred to the flask of an all-glass distillation apparatus having the delivery end of the condenser dipping into a 5 per cent. aqueous solution of potassium iodide. Ten ml. of sulphuric acid solution (6 N) were added through the funnel to the liquid in the distillation flask, and the liberated iodine was distilled over into the potassium iodide solution in the receiver. As the distillation proceeded N/10 sodium thiosulphate solution was added to the contents of the receiver, but a pale yellow colour, due to the presence of a little

iodine, was maintained. After the distillation and when the liquid in the distillation flask was perfectly colourless, the apparatus was disconnected, the condenser was washed down with a little potassium iodide solution, and the titration of the distillate was completed; a total of 23.52 ml. of *N*/10 sodium thiosulphate solution was recorded.

The iodine contained in the clear colourless liquid left in the distillation flask was then determined by the following modification of the method of Kolthoff and Yützy.⁴ The solution was neutralised with sodium hydroxide solution, methyl red being used as indicator, and then boiled down to 50 ml. and cooled. Fifteen g. of sodium chloride (AnalaR), 2 g. of sodium acid phosphate ($\text{NaH}_2\text{PO}_4 \cdot \text{H}_2\text{O}$, AnalaR), and finally 10 ml. of sodium hypochlorite solution (*N* in NaOCl -content and *N*/10 in NaOH) were added, and the liquid was heated to boiling. Ten ml. of sodium formate solution (30 g. of sodium hydroxide [pellets from sodium] and 32 ml. of formic acid solution [AnalaR, 90 per cent.] in 100 ml.) were added to the hot solution, precautions being taken to ensure that excess of sodium hypochlorite was decomposed by thorough washing of the sides of the beaker with water.

TABLE II

| Expt. No. | Alkali used Kind | Equiv. to <i>N</i> /10 ml. | Vol. of solution refluxed ml. | Iodine used g. | Duration of re- fluxing minutes | Result as <i>N</i> /10 alkali | |
|-----------|-----------------------------|----------------------------------|--|----------------------|--|-------------------------------|---|
| | | | | | | Smith's method ml. | Haslam and Roberts' method ml. |
| 14 | Sodium hydroxide | 25.00 | 120-180 | 0.5 | 30 | 23.5 | 25.01 |
| 15 | Sodium carbonate | 25.00 | " | " | 120 | 22.95 | 24.97 |
| 16 | Sodium bicarbonate | 25.00 | " | " | 180 | 23.2 | 24.95 |
| 17 | Sodium silicate | 24.90 | | | 90 | 23.32 | 24.82 |
| 18 | Sodium borate | 25.07 | | | 30 | 23.7 | 25.01 |
| 19 | Sodium aluminate | 25.00 | | | 90 | — | 25.06 |
| 20 | Trisodium phosphate | 24.83 | | | 90 | 21.5 | 23.69 |
| 21 | Disodium hydrogen phosphate | 25.20 | | | 90 | 21.0 | 22.85 |

Notes.—The sodium aluminate was prepared by dissolving 0.05 gm. of pure aluminium in 25 ml. *N*/10 sodium hydroxide solution, filtering into a flask and boiling for 2 minutes to remove any dissolved hydrogen before proceeding with the determination.

In the experiments with the sodium phosphates, a clear colourless liquid was not obtained on removal of the excess iodine by boiling. A yellow colour persisted even after prolonged boiling.

After standing for 10 minutes the solution was cooled in running water, diluted to 300 ml. and treated with 2 g. of potassium iodide, 50 ml. of sulphuric acid (3*M*) and 1 drop of ammonium molybdate solution (2.9 g. of $[\text{NH}_4]_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$ in 100 ml.). The liberated iodine was then titrated with *N*/10 sodium thiosulphate solution. A titration figure of 7.82 ml. of *N*/10 sodium thiosulphate solution (after correction for the result of a control experiment) was obtained.

This indicated that $\frac{7.82}{6}$ ml., i.e. 1.30 ml., of *N*/10 iodine was still present in the

clear colourless liquid in the distillation flask after complete removal by distillation of the iodine liberated on addition of sulphuric acid to the original alkali-iodine digestion product. The total amount of iodine found thus corresponded with 24.82 ml. of *N*/10 sodium hydroxide solution. The evidence also suggested that for every atom of sodium present as alkali in the original solution, one atom of iodine was present either in the form of sodium iodide or sodium iodate in the final reaction product. Hence it seemed that our difficulties in the application of Smith's method would be overcome by determining the total iodine in the final product of interaction of alkali and iodine, and not, as in Smith's method, the iodate alone.

Accordingly, various alkali solutions were taken and treated (a) by Smith's method, using 0.5 g. of iodine, the volume of solution refluxed being 120–180 ml., and (b) as in (a) up to the point where the excess of iodine not required in the reaction had been removed, after which the total iodine in the liquid remaining in the flask was determined as follows:—The solution in the flask was transferred to a graduated flask (200 ml.) and made to the mark with water. Fifty ml. of the solution were treated with 2 g. of sodium acid phosphate, and then immediately afterwards with 10 ml. of *N* sodium hypochlorite solution (see p. 401) and 15 g. of sodium chloride (Analar), and oxidised as described above, a final correction being applied as the result of a control experiment. The thiosulphate solution was standardised against potassium iodide; 50 ml. of a solution of potassium iodide containing 0.4151 g. of recrystallised potassium iodide per 200 ml. were oxidised to iodate by the hypochlorite procedure as a preliminary step.

The results in Table II are typical of those obtained.

The somewhat low results obtained with the sodium phosphates are not surprising; they are doubtless due to the influence of sodium acid phosphate, produced in the reaction, on the iodate-iodide mixture.

Our modification of Smith's method has proved to be of practical value in the determination of alkali in dark-coloured deposits and sludges containing appreciable proportions of sodium aluminate and silicate, *i.e.* for determinations in which ordinary titration methods with the use of indicators are not readily applicable.

Further, certain proprietary washing mixtures contain added dyestuff and, although as a rule interference due to dyestuff may be avoided by a preliminary treatment with sodium hypochlorite and hydrogen peroxide, followed by direct titration of alkali with the use of methyl orange as indicator, the alkali in such mixtures can invariably be determined by our modification, except when appreciable proportions of sodium phosphate are present.

Table III gives results obtained by Smith's method and by our modification of it in the analysis of solutions of other alkalis.

TABLE III

| Alkali used | | Result as <i>N</i> /10 alkali | | | | | |
|-------------|---------------------|-------------------------------|-------------------------------|----------------|-------------------------------|--------------------|--------------------------------|
| Expt. No. | Kind | Equiv. to <i>N</i> /10 ml. | Vol. of solution refluxed ml. | Iodine used g. | Duration of refluxing minutes | Smith's method ml. | Haslam and Roberts' method ml. |
| 22 | Potassium hydroxide | 24.14 | 120–180 | 0.5 | 30 | 22.9 | 24.12 |
| 23 | Potassium carbonate | 25.25 | " | " | 150 | 23.35 | 25.24 |
| 24 | Calcium hydroxide | 25.20 | " | " | 30 | 24.0 | 25.22 |
| 25 | Barium hydroxide | 24.75 | " | " | 30 | 20.12 | 24.3 |
| 26 | Barium hydroxide | 25.70 | " | " | 60 | — | 25.71 |
| 27 | Calcium carbonate | 24.85 | " | " | 150 | 22.25 | 24.53 |
| 28 | Barium carbonate | 24.93 | " | " | 150 | 21.9 | 24.29 |
| 29 | Ammonium carbonate | 24.45 | " | " | 30 | 13.63 | 19.36 |

Notes.—The low results with barium hydroxide are probably due to the difficulty of removing the very insoluble barium iodate from the reaction vessel.

In Expt. 26 the barium iodate was reduced to iodide by treatment with sulphurous acid prior to removal from the reaction vessel, in order to ensure that the whole of the combined iodine was determined in the subsequent hypochlorite oxidation.

In Expt. 29, after removal of most of the excess of iodine by boiling, a yellow colour remained and persisted even after prolonged boiling.

The results in Table III indicate that our modified method may be extended to the determination of alkali in solutions of potassium hydroxide, potassium carbonate, calcium hydroxide and barium hydroxide (provided that precautions are taken about the barium iodate produced in the reaction).

The results obtained with barium and calcium carbonates are not quite so trustworthy, and, as was to be expected, the reaction between iodine and ammonium carbonate does not proceed on the same lines as that between iodine and alkalis such as sodium hydroxide or potassium hydroxide.

REFERENCES

1. G. S. Smith, *ANALYST*, 1937, **62**, 590.
2. C. F. Walker and D. H. M. Gillespie, *Z. Anorg. Chem.*, 1899, **19**, 194; *Abst.*, *ANALYST*, 1899, **24**, 80.
3. E. Hinks, Presidential Address, *ANALYST*, 1930, **55**, 238.
4. I. M. Kolthoff and H. Yutzy, *Ind. Eng. Chem., Anal. Ed.*, 1937, **9**, 75.

RESEARCH DEPARTMENT

IMPERIAL CHEMICAL INDUSTRIES (ALKALI), LTD.

NORTHWICH

October, 1939

The Estimation of Lead in Drinking Water

BY C. H. MANLEY, M.A., F.I.C.

(Read at the Meeting of the North of England Section, January 27th, 1940)

THE tintometric estimation of lead in colourless waters presents no particular difficulties to the analyst. To a convenient quantity of the water, for example 25 or 50 ml., contained in a Nessler cylinder and made alkaline with ammonia, are added in succession ammonium citrate, potassium cyanide and sodium sulphide solution, and the colour produced is compared either with that given by a similarly treated standard lead solution or with a set of standard glass discs tinted to represent the colours yielded by different amounts of lead as lead sulphide.

On the other hand, waters which are naturally coloured present an obvious difficulty, especially when it is desired to exercise a close supervision of, say, a municipal supply containing very small concentrations of lead.

Examples of such waters are to be met with in many parts of the West Riding of Yorkshire, where, by reason of their peaty nature, they have a colour ranging from yellow to brown, and leave on evaporation a brown residue.

Thresh¹ called attention to the interference caused by the colour of moorland waters when he attempted to apply to them the method he had found successful for colourless waters, *viz.* addition of an acetic acid solution of gelatin followed by freshly prepared hydrogen sulphide² water.

Reith and de Beus³ attempted to overcome the difficulty by evaporating 100 ml. of water to about 50 ml. with 5 ml. of 5 per cent. ammonium persulphate solution, and treating it at 50° C. with 4 drops of 10 per cent. potassium cyanide solution and 10 ml. of 20 per cent. Rochelle salt solution. There was always the possibility, however, that at this stage filtration might be necessary before a clear liquid could be obtained. If so, this involved subsequent washing of the filter-paper with dilute hydrochloric acid to remove any adsorbed lead, before the addition of ammoniacal ammonium chloride and sodium sulphide could be made. A method followed for some time in my laboratory consisted in sulphating the ignited residue from 200 ml. of the water previously evaporated in a platinum basin, heating the residue to expel the excess of sulphuric acid, and extracting it with 40 per cent. ammonium acetate solution, after which the liquid was filtered and treated with ammonia, potassium cyanide, and sodium sulphide.

There seemed reason, however, to suspect that some loss of lead by volatilisation of lead monoxide might be occurring during the ignition of the total dissolved solids. Consequently, a separate volume of 500 ml. of the water was acidified at the outset with dilute sulphuric acid, and evaporated for the lead test alone, so that the lead should be present as the non-volatile sulphate throughout. Even so, on occasion the final tint was not always truly characteristic of lead sulphide, and it was suspected that traces of platinum might be interfering.

It was obvious that either the Allport and Skrimshire method⁴ or the S.P.A. method for the determination of lead in food-colouring materials⁵ would have enabled an accurate estimation to have been carried out, but both methods are too lengthy for routine testing purposes.

Hence, about a year ago a short-cut modification of the S.P.A. method (*loc. cit.*) was tried with highly successful results, and this is the method in use to-day. It is really the Direct Sulphate Precipitation Method which the S.P.A. Sub-Committee found inapplicable to the determination of lead in food-colouring materials on account of the presence of appreciable amounts of inorganic salts (chiefly sodium and ammonium sulphates) and of the interference of ferric sulphate, frequently likely to be present. The principle involved is that of wet oxidation followed by alcoholic precipitation of lead sulphate and solution of the latter in ammonium acetate prior to tinting. The intermediate stage involving precipitation of the lead as sulphide is thus dispensed with. This indeed should be possible, because the peaty moorland waters dealt with rarely contain more than 10 parts of dissolved solids per 100,000. If 8 of these are considered present as CaSO_4 and 500 ml. of the water are taken, this is equivalent to a CaSO_4 -content of 40 mg. in the 20 ml. of distilled water which is mixed with the alcohol used to complete the precipitation. This is the weight of CaSO_4 necessary to yield a saturated aqueous solution at 20° C.

METHOD.—A convenient quantity of the water (500 ml. when lead is likely either to be absent or at most present to the extent of 1/100 grain per gallon, *i.e.* 0.14 part per million) is mixed with 1 ml. of pure sulphuric acid and boiled down to a small bulk in a high-resistance globular glass flask. It is then transferred to a similar flask of 100-ml. capacity and the evaporation is continued until charring begins. The organic matter is then oxidised by heating with 1 ml. of conc. nitric acid, and the colourless solution resulting is twice evaporated with water until white fumes appear, before completing the precipitation of the lead sulphate overnight with a mixture of 20 ml. of water and 10 ml. of 95 per cent. alcohol. The precipitate is next transferred to a Gooch crucible containing prepared filter-paper pulp, and washed with two successive portions of 5 ml. of a mixture of water, alcohol and sulphuric acid in the proportions 20 : 10 : 1 by volume before being treated with two separate quantities of 10 ml. of hot 40 per cent. ammonium acetate solution. The pulp is washed twice with 5 ml. of hot water, and the filtrate is treated in a 50-ml. Nessler cylinder with 2 ml. of 8 per cent. ammonia, 0.02 g. of ammonium citrate, 0.02 g. of potassium cyanide, and 2 ml. of 10 per cent. sodium sulphide solution. The mixture is made up to 50 ml., and the lead is estimated either by comparison with a standard lead solution, each ml. of which contains 0.01 mg. of Pb, or in a B.D.H. Lovibond Nessleriser, using standard discs representing weights of lead ranging from 10γ–100γ. A blank estimation should also be carried out on the reagents. This is likely to give a correction of the order of 15γ.

With the foregoing method it has been found possible to recover quantitatively 50γ of lead added as nitrate both in absence and in presence of 500 ml. of Leeds City water, previously found free from lead.

I should like to express my thanks to Major A. Houlbrooke and Mr. H. Lobley for the assistance they have given me in connection with the present investigation.

REFERENCES

1. J. C. Thresh, *ANALYST*, 1924, **49**, 124.
2. ——— *id.*, 1921, **46**, 272.
3. J. F. Reith and J. de Beus, *Z. anal. Chem.*, 1935, **103**, 13; *Abst.*, *ANALYST*, 1935, **60**, 836.
4. N. L. Allport and G. H. Skrimshire, *ANALYST*, 1932, **57**, 440.
5. Second Report of the Sub-Committee on the Determination of Arsenic, Lead, and other Poisonous Metals in Food-colouring Materials to the Analytical Methods Committee of the Society of Public Analysts, *ANALYST*, 1935, **60**, 541.

CITY ANALYST'S LABORATORY
1, SWINEGATE, LEEDS, 1

February 23rd, 1940

Micro-Tests for Elements in Organic Compounds, Part II. Phosphorus, Arsenic and Antimony

By CECIL L. WILSON, M.Sc., Ph.D., A.I.C.

THE detection of phosphorus, arsenic and antimony in organic compounds depends on the conversion of these elements into corresponding inorganic anions.

Fusions on the macro scale may be carried out with (i) sodium nitrate and potassium nitrate, or (ii) sodium peroxide. On the micro scale very small amounts of material may be lost when (i) is used, while (ii) gives too violent a reaction. The lime fusion method of Feigl¹ gives insoluble compounds, which are not very suitable for subsequent identification.

The most satisfactory fusion mixture tested was one part of sodium peroxide intimately mixed with two parts of potassium nitrate. A few crystals of the sample are mixed with about three times their weight of this, and the mixture is filled into a bulb (4 mm. in diameter), blown on the end of a thin-walled glass tube, 7 cm. \times 2 mm. The fusion mixture alone is placed on top of this until the bulb is half full, and its contents are fused in such a way as to melt slowly. After the fusion, the bulb is heated strongly and dipped into a large drop of water on a microscope slide or a small watch-glass, or, preferably, in a 1-ml. crucible. The mass is ground and stirred thoroughly, and the resultant solution or suspension is tested for the appropriate anions.

PROCEDURE.—Antimony and arsenic are first tested for by a modified Gutzeit test. Of the many recommended methods for the separate identification of arsine and stibine, the only one that proved completely successful on the micro scale depended on reduction with tin and hydrochloric acid in presence of platinum.

A "couple" is made from a strip of platinum foil, 20 \times 2 mm., and a strip of tin foil, 60 \times 2 mm. The strip of tin is twisted loosely and then wound from one end of the platinum strip to the other in such a way as to leave a considerable area of bright platinum visible. The platinum strip is in turn wound about its short edge into a loose flat spiral of such a size as to drop into the reaction vessel of the gas detection apparatus described elsewhere (see p. 407). A drop (one-half) of the fusion *suspension* is transferred to the chamber. (Under the conditions of the fusion described, antimony appears to form an insoluble salt, probably sodium pyroantimoniate.) Five drops of 5 *N* hydrochloric acid are added, and heating is begun.

Arsenic.—By this treatment, arsenate is reduced to arsine, which rapidly blackens the silver nitrate test-paper. If antimony is present, it is retained. There may, under continued reduction, be a very faint yellowing of the test-paper if antimony is present and arsenic absent, but, since even minute traces of arsenic darken the paper appreciably, this may be neglected.

Antimony.—In the course of the reduction with the tin-platinum "couple," antimony is deposited as the metal, in the form of a black coating on both tin and platinum. This is sufficient verification of its presence (dulling of the platinum by small amounts of antimony is very easily seen) but, if desired, confirmation may be obtained by removing the "couple," separating the tin and platinum and washing the platinum strip thoroughly with air-free distilled water. The antimony is then dissolved off the platinum in the reaction chamber of the gas detection apparatus by heating gently with three drops of conc. hydrochloric acid. When the platinum is completely cleared it is removed by means of a fine glass rod. Several drops of water and a few chips of zinc are added, and the presence of antimony will be shown by the blackening of silver nitrate test-paper.

Phosphorus.—If arsenic is absent, a direct test for phosphorus is made in the remainder of the fusion liquid. The clear liquid is transferred to a slide and rendered just acid with sulphuric acid. The reagent solution is made up as follows: 3 g. of ammonium acetate and 10 ml. of ammonia (sp.gr. 0.88) are made up to 25 ml. with water. After complete solution has taken place, 1 g. of magnesium acetate is dissolved. A large drop of this reagent is added to the test drop, so that the whole has a *strong* odour of ammonia.*

The drop is allowed to stand for a few minutes, and if only small or poorly developed crystals have appeared by then, the whole is heated just to boiling, and allowed to cool. The presence of phosphorus (as phosphate) is indicated by the formation of characteristic crystals, as described by Chamot and Mason.³

If arsenic is present, it must first be removed. The test-drop is placed in the reaction chamber, and to it are added five drops of 5 *N* hydrochloric acid and a few chips of zinc. Reduction is continued until no arsine can be detected on addition of a further drop of acid and a small chip of zinc. A large drop of the liquid is then removed, and tested for phosphate as before. Under these conditions the forms of the phosphate crystals tend to be less feathery than those from an unreduced phosphate solution.

The following compounds were satisfactorily used for the tests: Sodium glycerophosphate, sodium phenyl phosphate, triphenyl phosphate, nucleic acid; sodium cacodylate, tryparsamide, *p*-arsanilic acid, sodium methyl arsonate; antimony strontium tartrate, antimony potassium tartrate, antimony oxalate.

The sensitivity of the tests varied with the compounds, but in general, in mixtures containing 5 to 20% of phosphorus compound, 10 to 20% of arsenic compound and 20 to 30% of antimony compound, the elements could be detected correctly. Although these figures are not so low as those given by Feigl,⁴ they apply to mixtures, for which his methods are not suitable.

SUMMARY.—The detection of phosphorus, arsenic and antimony micro-chemically in organic mixtures is preceded by oxidation to the corresponding inorganic anions. Phosphorus is detected by its characteristic double magnesium ammonium compound. To distinguish arsenic and antimony, reduction is effected by a tin-platinum "couple," and a modified Gutzeit test is applied.

I wish to express my indebtedness to The British Drug Houses, Ltd., who kindly supplied me with samples of organic compounds.

REFERENCES

1. "Qualitative Analysis by Spot Tests," F. Feigl, translated by J. W. Matthews, 1937, p. 246.
2. "Lehrbuch der Mikrochemie," F. Emich, 1926, 148, 180. "Handbook of Chemical Microscopy," E. M. Chamot and C. W. Mason, II, 1931, 313.
3. E. M. Chamot and C. W. Mason, *loc. cit.*, 77, 113.
4. F. Feigl, *loc. cit.*, 245, 246.

THE SIR JOHN CASS TECHNICAL INSTITUTE
LONDON, E.C.3

February, 1940

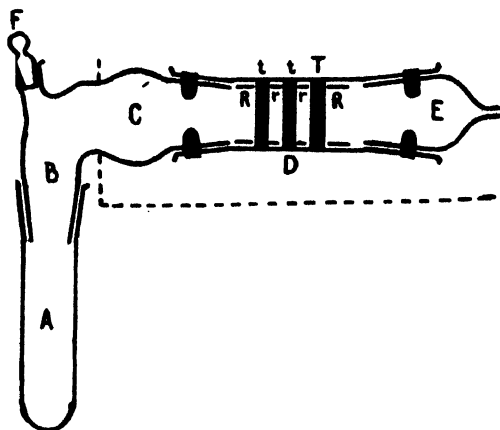
* Since it is essential to maintain the relative proportions of ammonia and the two acetates, particularly when small amounts are being tested for, it is preferable to use this compound reagent solution, rather than to add the reagents separately, as described by other authors.*

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

A GAS DETECTION APPARATUS FOR QUALITATIVE MICROCHEMISTRY

THIS apparatus, originally devised for the detection of arsine or stibine in presence of hydrogen sulphide, *cf.* p. 405, should prove useful whenever it is necessary to filter off one gas before testing for another.



The reaction vessel, A, shown in the diagram, is a fusion tube or micro-test-tube, connected with an arm, B, which leads the gases to the testing chamber and through which reagents may be added. A bulb, C, in this arm contains cotton-wool or glass-wool, to retain spray. The testing chamber, D, is a length of straight tubing. Within this is a series of glass rings, R, r, which just slide in it; these may be prepared by cutting them from another piece of glass tubing of the appropriate size. These serve to hold apart and in position test-papers, t, T, impregnated with suitable reagents. The test-papers are discs cut from "spot" paper by a cork borer, and they should fit the walls of the testing chamber tightly at every point. A spare testing chamber is necessary when a blank test has to be applied to the reagents. At the end of the apparatus is a piece of tubing, E, drawn out to a fine aperture to avoid undue contamination of the test-papers with the atmosphere. The whole apparatus is mounted by means of two metal spring clips on to a piece of asbestos board, which may be held in a retort stand for the reaction vessel.

In testing *e.g.* for arsine in presence of hydrogen sulphide, the two testing chambers are fitted as follows:—The chamber is held vertically with the lower end blocked. The first stop ring, R, is placed in position, and a disc of paper, t, is pressed home with a plunger (conveniently a glass tube of the same diameter as the rings). A large drop of lead acetate solution is applied by means of a capillary tube to the disc, which must be completely wet by the liquid. One of the intermediate rings, r, is then inserted, followed by a second disc of paper. This is also impregnated with lead acetate solution, and followed by another intermediate ring and a third disc of paper, T. Unless a very small trace of arsine in presence of a very large amount of hydrogen sulphide is to be expected, no more filtering test-papers are required, and this third disc is impregnated with a small drop of dilute nitric acid and a drop of silver nitrate solution. The second stop ring is then added.

Two testing chambers are prepared in this way. One is fitted into the apparatus. The ends of the other are plugged until it is required.

Several small chips of zinc and then five drops of 5 N hydrochloric acid are introduced through the filling aperture, F, and the apparatus is heated so that vigorous effervescence takes place for a fixed time, say, two minutes. When very small traces of arsenic are being sought for, this period must be longer.

The testing chamber is replaced by the spare chamber, and the test material is introduced through F. Heating (and effervescence) are continued for the same length of time, after which the chamber is withdrawn, and the discs of paper are removed and examined.

Those from the blank test should, of course, be white, or nearly so. Of the discs in the second test chamber, No. 1 may be black or brown, showing the presence of hydrogen sulphide. No. 2 should be white, or nearly so. If No. 3 is darker in colour than No. 2, and also than No. 3

of the blank, then arsine (or stibine, when no other method of distinguishing between the two has been used) is present.

If disc No. 2 is appreciably darkened, the test must be repeated with more filtering discs present. The length of the testing chamber allows for the addition of more discs, and in the final test just sufficient should be present to ensure that the final one immediately preceding the actual test-disc is unaffected.

After considerable experience with the apparatus, it will be found that it allows not only of detection of the gases, but also of a rough estimation of the relative amounts present.

C. L. WILSON

THE SIR JOHN CASS TECHNICAL INSTITUTE
JEWRY STREET, ALDGATE, E.C.3

October, 1939

THE DETERMINATION OF COCOA ALKALOIDS IN CONFECTIONERY

THE process here described is concerned with the final extraction of the alkaloids after treatment of the material according to the method of Moir and Hinks (ANALYST, 1935, 60, 439). In that method 10 to 20 g. of confectionery are heated under reflux with 200 ml. of 80 per cent. alcohol and 2.5 g. of magnesia for 1½ hours and then filtered through a Buchner funnel. The residue is extracted with two further quantities of 80 per cent. alcohol, and the extract is filtered. The filtrate is evaporated to low bulk, and water is added to bring the volume up to about 100 ml. This solution is slightly acidified, transferred to a 200-ml. flask, treated with 5 ml. of zinc acetate solution followed by 5 ml. of potassium ferrocyanide solution, mixed, made up to volume and filtered, and a suitable portion of the filtrate is concentrated to about 10 ml.

In the original method this concentrate is extracted with chloroform, and, if 20 to 30 mg. of alkaloids are present, as many as ten extractions are required to remove them completely (*cf.* A. E. and H. A. Parkes, ANALYST, 1937, 62, 791), and the resulting alkaloids are invariably impure, thus necessitating a nitrogen determination. With the idea of avoiding this tedious extraction process I investigated the possibility of converting the small quantities of theobromine into caffeine (which is much more soluble) in the presence of many times its weight of sugar.

In a preliminary experiment 10 mg. of pure theobromine and 5 g. of sugar were dissolved in about 10 ml. of water, and the solution was transferred to a small separating funnel, and treated with 1 ml. of *N* sodium hydroxide solution, followed by 0.5 ml. of methyl sulphate, the separator being shaken occasionally during 5 minutes. One ml. of 50 per cent. sodium hydroxide solution was then added to decompose the excess of methyl sulphate and, after standing for 10 minutes, the mixture was extracted with three successive portions of 30 ml. of chloroform, and the chloroform extracts were washed once with water and run through a dry filter-paper into a weighed flask. The chloroform was evaporated and the residue was dried at 100° C. and weighed. It consisted of white crystals of caffeine weighing 10.6 mg.

Ten g. of home-made chocolate cake were treated according to the method of Moir and Hinks as described, the final concentrate was made up to 20 ml. and 10 ml. were extracted directly with chloroform.

| | |
|--|----------|
| Weight of alkaloid after 5 extractions, each with 30 ml. of chloroform | g. |
| " " " " 10 " " " " | = 0.021 |
| Nitrogen calculated as caffeine " " " " | = 0.024 |
| " " " " " " " " | = 0.0156 |

The alkaloids recovered by this process were amorphous and darkened considerably when treated with sulphuric acid.

The other 10 ml. were treated with methyl sulphate as described above.

| | |
|--|---------|
| Weight of alkaloid after 3 extractions, each with 30 ml. of chloroform | g. |
| " " " " 5 " " " " | = 0.015 |
| " " " " " " " " | = 0.015 |

The residue from this chloroform extract was white and crystalline and melted at 234° C., which corresponds with pure caffeine.

CONCLUSION.—After treatment of cocoa confectionery according to the method of Moir and Hinks the alkaloids are most conveniently recovered and weighed as pure caffeine by treating the clarified extract with methyl sulphate as described above. The weight of caffeine multiplied by 0.928 corresponds with the weight of the original alkaloids as theobromine, and an average factor for converting caffeine into mixed alkaloids is 0.935.

COUNTY ANALYST'S OFFICE

BRADFORD

J. C. HARRAL
May, 1940

THE IODINE VALUE OF SOFT PARAFFINS

As the iodine value of a sample of soft paraffin may give some indication of its liability or otherwise to become overheated during the preparation of bleach ointment, and may also affect the rate of the loss of available chlorine (*cf.* Macmorran, p. 423), it is necessary to use a method of determining it that will give concordant results in the hands of different workers. The B.P. method, which is a modification of the Wijs method, is unsuitable because soft paraffins are largely insoluble at ordinary temperature in the mixture of carbon tetrachloride and reagent as ordinarily used. Iodine values of soft paraffins have been published by Bryant and Spence¹ and by Fishburn,² but the method used is not stated. It was found that Rosenmund and Kuhnenn's pyridine sulphate bromide method did not offer any advantage over the more commonly used Wijs method.

A study of the conditions showed that solution of the paraffin was greatly improved by reducing the volumes of the paraffin and the reagent. By using 1 g. of the sample, 20 ml. of carbon tetrachloride and 10 ml. of the official Wijs reagent, a clear solution was obtained with 35 different samples of white and yellow soft paraffins, and only with 2 samples, probably containing cerasine, was there a very slight precipitate. By varying the time of standing and excess of reagent it was found that, after an initial fairly rapid absorption of iodine, combination continues slowly for a very long time. Thus the following iodine values were obtained with a typical sample of B.P. yellow soft paraffin (1 g. with 20 ml. of carbon tetrachloride and 10 ml. of Wijs reagent):—After half-an-hour, 9.8; 1 hour, 10.0; 1½ hours, 10.8; 2 hours, 11.26; 72 hours, 12.7. In further experiments with 0.5 g. of the sample and 5 ml. of Wijs reagent, similar, but slightly lower, results were obtained. Raising the temperature or increasing the excess of reagent resulted in higher values, as was to be expected.

The results of these experiments show that it is necessary to fix a suitable time of contact with the reagent. On plotting iodine absorption against time, the curve with some samples showed a somewhat sharp rise at 1 hour, but became considerably flatter before the 1½ hour period. For this reason it was considered preferable to increase the usual 1 hour to 1½ hours. The following procedure, based on experiments with 37 representative samples, is recommended. It gives results agreeing within ± 1 per cent. (usually ± 0.5 per cent.) in duplicate determinations.

METHOD.—An amount between 0.95 and 1.05 g. of the sample, accurately weighed, is dissolved with the aid of gentle heat in 20 ml. of carbon tetrachloride in a glass-stoppered flask. The solution is cooled to between 19° and 21° C., and treated with 10 ml. of Wijs solution. The flask is closed and left for 1½ hours at 19° to 21° C. Ten ml. of 10 per cent. potassium iodide solution and 100 ml. of water are then added, and the excess of iodine is titrated with *N*/10 thiosulphate solution. A blank test is made without the soft paraffin.

By this method 18 samples of yellow soft paraffin, obtained mainly from importers, and of as varied a character as possible, gave iodine values ranging from 2.5 to 12.3 with an average of 9.6. Thirteen samples gave results between 9.5 and 11.5.

With 19 samples of the white variety, of similar origin, the iodine values ranged from 0.9 to 11.4 with an average of 5.1. Seven samples gave figures between 4.5 and 5.5.

As a rule, there was a close relationship between the iodine values of soft paraffins and their liability to become over-heated in the preparation of bleach ointment, the higher the iodine value, the greater the tendency towards over-heating. There were, however, one or two anomalous samples; the thermal bromine method was tried to see if it would give an indication of abnormality, but the results were practically parallel with the iodine values.

Other characteristics of soft paraffins were described in a previous communication.³

H. BRINDLE

REFERENCES

1. E. G. Bryant and J. Spence, *Year Book of Pharmacy*, 1926, 445.
2. A. G. Fishburn, *Pharm. J.*, 1940, 144, 35.
3. H. Brindle, *Quart. J. Pharm.*, 1939, 12, 361.

PHARMACY DEPARTMENT
MANCHESTER UNIVERSITY

March 19th, 1940

DETERMINATION OF CHROMATES IN SEWAGE LIQUORS

OWING to the increased use of chromic acid and chromates in industry the presence of chromate in sewage liquors is now of fairly common occurrence. If any significant quantity is present, the usual determinations of organic matter and B.O.D. are materially affected, and it is necessary to make a correction for the iodine liberated by the chromate from the potassium iodide used in these tests. Jenkins and Hewitt¹ determined directly the chromate correction to be used when applying the 4-hours oxygen-absorbed test to crude sewage; they acidified the sample in the presence of excess of potassium iodide and titrated the iodine liberated by interaction of chromate and iodide. Although simple, this method is not very reliable, since the iodine liberated by nitrites is also included in the correction. Nitrites are occasionally present in crude sewage and are of frequent occurrence in purified sewage. I have also found that the result so obtained

may vary with the age of the sample. Spencer's method,* in which the organic matter is first oxidised by alkaline permanganate, has an indefinite end-point, and has also been found to give erratic results.

Illustrations of these defects became available when, owing to an accident, about 15 cwt. of sodium dichromate was discharged into a sewer. This was accompanied by a quantity of a lead solution which caused partial precipitation of the chromate in the sewer and subsequent removal in the settling tanks at the sewage works. The following results were obtained with 24-hour samples taken at the sewage works:

| Sample of: | Chromium, parts per 100,000 found | |
|---|-----------------------------------|--------------------------------|
| | By Spencer's method | By Jenkins and Hewitt's method |
| Crude sewage (believed to contain 0.4-0.5 part of Cr per 100,000) | 0.83 | 0.65 |
| Settled sewage | 1.48 | 1.52 |
| Partly purified sewage | 0.96 | 1.22 |
| Final effluent | 2.08 | 1.39 |

These figures have not been corrected for the presence of nitrites. The sample of crude sewage was again examined by Spencer's method, but was left to stand overnight after the excess alcohol had been boiled off. When potassium iodide was added to the acidified solution, this appeared to contain 59.4 parts of chromium per 100,000.

The following method was then tried:—Fifty ml. of the sample were measured into a stoppered bottle and a few crystals of sodium azide were added. The mixture was acidified with a few ml. of conc. phosphoric acid, rotated, and left to stand for several minutes. Excess of potassium iodide was then added, followed by 5 ml. of 25 per cent. sulphuric acid, and the liberated iodine was titrated with $N/40$ thiosulphate solution.

Comparison of the results obtained by this method with those obtained by the direct method on two further samples gave:

| Sample of: | Chromium,* parts per 100,000, found | |
|---|-------------------------------------|---------------|
| | Azide method | Direct method |
| Partly purified sewage | 0.20 | 1.00 |
| Final effluent | 0.35 | 2.29 |
| " " (after 5 days' incubation at 80° F.) .. | 0.35 | 0.87 |

* Exact chromium content of liquors unknown.

In order to test the effectiveness of this method for the destruction of nitrites, quantities of sodium nitrite solution were added to a number of portions of $N/4000$ sodium dichromate solution so as to give a range of concentration up to 0.04 part of nitrous nitrogen per 100,000. The chromium-content was determined by the direct method and by the azide method. It was found that the result obtained by the azide method was independent of the nitrite concentration and that good end-points were obtained. An amount of 0.35 part of chromium per 100,000 in the final effluent from the works, with a total flow for the period of 12 million gallons, is equivalent to a total of about 10½ cwt. of sodium dichromate. Having regard to the precipitation which must have taken place and the further "loss" in the settling tanks, this figure is in fairly good agreement with the 15 cwt. known to have been discharged.

The azide-iodine reaction of Feigl³ does not interfere, owing to the high acidity of the mixture.

REFERENCES

1. S. H. Jenkins and C. H. Hewitt, *J. Soc. Chem. Ind.*, 1940, 49, 41.
2. J. H. Spencer, Ann. Summer Conf., Inst. Sewage Purif., July, 1939 (see *Sewage Purification*, 1939, 1, 356).
3. F. Feigl, *Z. anal. Chem.*, 1928, 74, 369.

THE LABORATORY
STOCKPORT SEWAGE WORKS

D. DICKINSON
May, 1940

The following notification (dated June 6th, 1940) of Amendment of the List of Public Analysts appointed by Local Authorities with the approval of the Minister of Health has been made:

| Authority | | | | | Public Analyst |
|---------------------------------------|----|----|----|--|--------------------------------|
| Castleford Urban District | .. | .. | .. | | F. W. M. JAFFÉ |
| Halifax County Borough | | | | | R. MALLINDER |
| Stockton-on-Tees Borough | | | | | C. J. H. STOCK |
| Thurrock Urban District | | | | | A. W. STEWART |
| Hereford Borough | | | | | F. G. D. CHALMERS (Additional) |
| Burton-upon-Trent County Borough . | | | | | R. MALLINDER |
| Morecambe and Heysham Borough . | | | | | J. R. STUBBS |
| Kingston-upon-Thames Borough . | | | | | E. HINKS |
| Batley Borough | | | | | F. W. M. JAFFÉ |
| Huddersfield County Borough .. . | | | | | R. MALLINDER |
| Chislehurst and Sidcup Urban District | | | | | F. W. F. ARNAUD |
| Brierley Hill Urban District .. . | | | | | E. V. JONES |
| " " " " " " " " | | | | | F. DIXON (Deputy) |
| Mitcham Borough | | | | | E. HINKS |
| " " " " " " " " | | | | | D. D. MOIR (Deputy) |
| Scarborough "Borough" | | | | | R. MALLINDER |
| Carshalton Urban District .. . | | | | | E. HINKS |
| " " " " " " " " | | | | | D. D. MOIR (Deputy) |
| Crosby Borough .. " " " " " " | | | | | W. H. ROBERTS |
| Gosport Borough | | | | | R. P. PAGE |
| Westmorland County Council .. . | | | | | W. H. ROBERTS |
| Kendal Borough | | | | | W. H. ROBERTS |
| Gillingham Borough | | | | | F. W. F. ARNAUD |
| Beddington and Wallington Borough . | | | | | H. A. WILLIAMS |
| Mansfield Borough | | | | | W. W. TAYLOR |
| Darwen Borough | | | | | J. R. STUBBS |
| " " " " " " " " | | | | | ARNOLD LEES (Deputy) |
| Ilford Borough | | | | | J. H. HAMENCE (Joint) |

THE Ministry of Agriculture and Fisheries has notified the Society of the following changes in the appointments of Agricultural Analysts and Deputy Agricultural Analysts which have taken place since June 28th. 1939.

W. F. GREEVES, F.I.C., Deputy Agricultural Analyst to the County Council of East Suffolk.

The Margarine (Addition of Borax) Order, 1940

WE are informed that the Ministry of Food has made it a condition of the licences issued to manufacturers under the Margarine (Addition of Borax) Order, 1940 (see p. 413), that the amount of borax added to margarine should not exceed 0.25 per cent. expressed as boric acid.

EDITOR

July 11th, 1940

Home Office

Poisons

STATUTORY RULES AND ORDERS. 1940. No. 452*

THE POISONS (AMENDMENT) RULES, 1940, DATED MARCH 29, 1940, MADE BY THE SECRETARY OF STATE UNDER SECTION 23 OF THE PHARMACY AND POISONS ACT, 1933 (23 & 24 GEO. 5. C. 25). The following Rules have been made:

1. In the First Schedule to the Poisons Rules, 1935, as amended by the Poisons (Amendment) Rules, 1937, and the Poisons (Amendment) Rules, 1938:
 - (a) in the item "Arsenical poisons" there shall be inserted at the end the words "and except dentifrices containing less than 0.5 per cent. of acetarsol";
 - (b) in the item "Para-aminobenzenesulphonamide" for the words "derivatives of para-aminobenzenesulphonamide having one or both of the hydrogen atoms of the para-amino group substituted by other radicals" there shall be substituted the words "derivatives of para-aminobenzenesulphonamide having any of the hydrogen atoms of the para-amino group or of the sulphonamide group substituted by another radical"; and
 - (c) there shall be inserted the following substances:—Sulphonal; alkyl sulphonals.
2. In Group II in the Third Schedule to the said Rules there shall be inserted in the first column the poison "Antimony, chlorides of" and opposite those words there shall be inserted in the second column the word "Polishes."
3. In the Fourth Schedule to the said Rules, in the item "Para-aminobenzenesulphonamide" for the words "derivatives of para-aminobenzenesulphonamide having one or both of the hydrogen atoms of the para-amino group substituted by other radicals" there shall be substituted the words "derivatives of para-aminobenzenesulphonamide having any of the hydrogen atoms of the para-amino group or of the sulphonamide group substituted by another radical."
- 4.—(1) These Rules may be cited as the Poisons (Amendment) Rules, 1940, and the Poisons Rules, 1935, the Poisons (Amendment) Rules, 1937, the Poisons (Amendment) Rules, 1938, and these Rules may be cited together as the Poisons Rules, 1935 to 1940.
- (2) These Rules shall come into operation on the first day of April, 1940.

HOME OFFICE,
WHITEHALL

JOHN ANDERSON,
One of His Majesty's Principal Secretaries of State.
29th March, 1940

STATUTORY RULES AND ORDERS. 1940. No. 453*

THE POISONS LIST (AMENDMENT) ORDER, 1940, DATED MARCH 29, 1940, MADE BY THE SECRETARY OF STATE UNDER SECTION 17 (5) OF THE PHARMACY AND POISONS ACT, 1933 (23 & 24 GEO. 5 C. 25), AMENDING THE LIST OF THE SUBSTANCES WHICH ARE TO BE TREATED AS POISONS FOR THE PURPOSES OF THAT ACT.

Whereas the Poisons Board has recommended to me that the list of the substances which are to be treated as poisons for the purposes of the Pharmacy and Poisons Act, 1933(a), should be amended so that certain additional substances should be included in Part I of the said list:

Now, therefore, in pursuance of Section 17 (5) of the Pharmacy and Poisons Act, 1933, I hereby order as follows:

1. In Part I of the Poisons List (b) in the item "Para-aminobenzenesulphonamide" for the words "derivatives of para-aminobenzenesulphonamide having one or both of the hydrogen atoms of the para-amino group substituted by other radicals" there shall be substituted the words "derivatives of para-aminobenzenesulphonamide having any of the hydrogen atoms of the para-amino group or of the sulphonamide group substituted by another radical."
2. This Order may be cited as the Poisons List (Amendment) Order, 1940, and shall come into operation on the first day of April, 1940.

HOME OFFICE
WHITEHALL

JOHN ANDERSON,
One of His Majesty's Principal Secretaries of State.
29th March, 1940

(a) 23 & 24 Geo. 5. c. 25.

(b) See Schedule to the Poisons List Confirmation Order, 1935 (S.R. & O. 1935 (No. 1238), p. 1383), as amended by the Poisons List (Amendment) Order, 1937 (S.R. & O. 1937 (No. 1029), p. 1917), and the Poisons List (Amendment) Order, 1938 (S.R. & O. 1938 (No. 1547), II, p. 2829).

Ministry of Food

STATUTORY RULES AND ORDERS. 1940. No. 982*

EMERGENCY POWERS (DEFENCE)

Food

THE MARGARINE (ADDITION OF BORAX) ORDER, 1940, DATED JUNE 14, 1940

IN exercise of the powers conferred on him by Regulation 55 of the Defence (General) Regulations, 1939^(a), and of all other powers him enabling the Minister of Food (hereinafter referred to as "the Minister") hereby makes the following Order:

1. In this Order "Borax" includes Boric Acid and Borates.
2. Subject to any directions given or except under and in accordance with the terms of a licence granted by or on behalf of the Minister no person shall:
 - (a) produce or manufacture for sale any margarine which contains any added borax; or
 - (b) treat with borax or add borax to any margarine.
 Provided that the provisions of this Article shall not apply to margarine which is intended to be exported or intended for use as ships' stores.
3. It shall be lawful for any person subject to the provisions of this Order and of any other Order made or directions given by the Minister:
 - (a) to produce or manufacture for sale or sell or offer or expose for sale or deposit in any place for the purposes of sale any margarine notwithstanding that such margarine contains added borax; and
 - (b) to produce or manufacture for sale or sell or offer or expose for sale any article of food other than margarine, which contains borax necessarily introduced by the use in the preparation of that article of margarine containing borax.
4. Infringements of this Order are offences against the Defence (General) Regulations, 1939
5. This Order may be cited as the Margarine (Addition of Borax) Order, 1940.

By Order of the Minister of Food.

H. L. FRENCH,
Secretary to the Ministry of Food.

Dated the 14th day of June, 1940.

Department of Scientific and Industrial Research

REPORT OF THE NATIONAL PHYSICAL LABORATORY FOR 1939†

IN the Report of the Executive Committee, the Director (Dr. C. G. Darwin) refers to the work of the Laboratory in war time. Although each of the three services has a strong research department, certain problems arising out of war conditions continue to be submitted, so that the Laboratory is again playing its part, although in less degree than in the last war. The policy of not dispersing the scientific staff has been fully justified, and has enabled the peace-time function of tendering advice and help to industry on scientific and technical problems to be fully maintained. Subjects of special interest to which reference is made in the Reports of the Superintendents of the Physics Department and Engineering Department include the following:

SPECIFIC HEAT AND MELTING-POINT OF VERY PURE IRON.—A specimen of iron (purity 99.99 per cent.), prepared in the Metallurgy Department, had a specific heat of 0.11 at 50° C., and melted at 1533° ± 5° C.

INTERNATIONAL TEMPERATURE SCALE.—In order to obtain a "fixed" point between the b.p. of sulphur (444.60° C.) and the freezing-point of gold (1063.0° C.), the b.p. of selenium (684.8 ± 0.1° C. for normal atmospheric pressure) was determined by means of calibrated platinum thermo-couples.

INTERNAL STRAIN IN METALS.—It was found by X-ray measurements that the atomic lattices of copper, silver, nickel, iron and molybdenum expand when the metal is subjected to stress or cold-worked; this represents an unstable condition, since, on continuing the cold-working, there may be a reversion to normal dimensions. It was also found that the breakdown of metallic grains into finer components proceeds to a limit which depends on the metal and the temperature. This lower limit, which is a new physical constant, was determined for each of the above-mentioned metals.

TOOTH STRUCTURE.—Earlier X-ray investigation showed that tooth enamel contains two oriented groups of apatite crystals, the hexagonal axes of which make angles of about 5° and 40° respectively with the prism direction. Further work has indicated that the 5° group usually

(a) S.R. & O. 1939, No. 927.

* H.M. Stationery Office, York House, Kingsway, London, W.C.2. Price 1d. net.

† H.M. Stationery Office, York House, Kingsway, London, W.C.2. 1940. Price 2s. 6d. net.

predominates within the prism, and the 40° group in the interprismatic substance. The degree of calcification may be indicated by the interference colours shown by enamel in polarised light. This method has confirmed the X-ray result that in the human tooth calcification diminishes inwards from the outer enamel surface. A method of determining the proportion of calcified matter in the enamel or dentine from point to point in a tooth section has been based on micro-photometric measurements of radiographs.

HAEMOGLOBINOMETER STANDARDISATION.—Colorimetric estimation of carboxyhaemoglobin in a haemoglobinometer tube has hitherto been based on comparison with a standard solution of carboxyhaemoglobin defined in terms of the oxygen-content of the haemoglobin. Haldane found that the blood of an average healthy person contained 18.5 ml. of oxygen per 100 ml., and this value was adopted in preparing colour tubes. A special Committee of the British Standards Institution decided that it would be preferable to define the standard solution colorimetrically. In order that there should be agreement with existing colour tubes, carboxyhaemoglobin, prepared by Haldane's method, was colorimetrically measured, and the result was taken as defining the standard solution. A sensitive colour comparator has also been devised to enable colour tubes to be accurately calibrated.

SURFACE FINISH OF METALS.—In a photoelectric instrument, designed in the Laboratory, the ratio of the regularly reflected light to the scattered light is measured; a high value denotes a high quality of surface finish. This instrument, which has been tested on numerous materials, could be constructed and used in industrial laboratories.

ELECTRICAL STANDARDS.—It has been proposed to change from international units to absolute units in 1940, and, with this end in view, standards of resistance and standard cells were calibrated in both systems and sent to the International Bureau of Weights and Measures for comparison with standards from other countries. It was found that all the values for the ohm (absolute) agreed within 2 parts per 100,000, and it was agreed that the most probable value is:

$$1 \text{ international ohm} = 1.00049 \text{ ohm (absolute).}$$

With regard to the ampere and volt, only the National Physical Laboratory and the National Bureau of Standards agreed within about 1 part in 100,000. The Consultative Committee was therefore unable to recommend a value with limits closer than 1 or 2 parts per 10,000, and therefore proposed the adoption for the present, of the values:

$$\begin{aligned} 1 \text{ international ohm} &= 1.0005 \text{ ohm (absolute)} \\ 1 \text{ international ampere} &= 0.9999 \text{ ampere (absolute)} \\ 1 \text{ international volt} &= 1.0004 \text{ volt (absolute)} \end{aligned}$$

Owing to the outbreak of war, no action is being taken at present to introduce the absolute units.

DIELECTRICS.—Previous work on the properties of plastics as electrical insulating materials had been mainly confined to synthetic resins of the phenolic type. A similar investigation of aniline resins and of pure hydrocarbon resins, including polystyrene and polyethylene, has shown that these materials have advantages for certain classes of electrical work. Experiments have also been made on the properties of laminated boards made from paper and the various resins.

INTERNATIONAL VISIBILITY CURVE.—Further work has been done on the curve relating the energy for each wavelength of light with its effect as perceived in the normal eye. It was agreed at the meeting of the International Commission in Illumination in July that no change should be made. A review of the work of the past few years has shown that changes in the conditions of observation, particularly field size and brightness, may cause considerable variations in the curve. There is even some evidence of variation with the season, possibly owing to differences in the vitamin A content of the normal diet.

INVESTIGATIONS RELATING TO AIR RAID PRECAUTIONS LIGHTING RESTRICTIONS.—The problem of measuring the intensities of very faint lights has been studied. For many purposes it is sufficient to ascertain if a light falls below some fixed standard, and a simple portable gauge to do this has been designed. The comparison field is provided by radium luminous compound, which does not require frequent renewal. By suitable modification the instrument can be used for measurement instead of gauging.

A simple photometer has been designed for measuring the brightness of materials intended to be luminescent after exposure to daylight or ultra-violet light.

In connection with the "black-out," the opacity of black textiles has been studied, and "complementary lighting" has been investigated. This method consists, for example, in using red or yellow light in a building and preventing it being visible by night by having windows of blue glass of sufficient transparency to allow enough daylight to enter during the day time. Delicate adjustment of the pairs of colours is necessary.

LUBRICATION RESEARCH.—An investigation of the comparative lubrication value of animal, vegetable and mineral oils has been completed by the Engineering Department. It was found that under all conditions, other than a complete fluid film, animal and vegetable oils are superior to mineral oils from the purely mechanical point of view. As a rule, no serious effects were caused by the chemical instability of fatty oils, and the temperature of journal bearings could be raised to 200° C. several times without apparent loss of lubricating quality of the oil. Advantages that fatty oils have over mineral oils are reduction of friction at high load and low speed, and, under extreme conditions, reduction of wear and increase in the breakdown load. Little difference was observed in the behaviour of vegetable and animal oils.

International Union of Chemistry

TENTH REPORT OF THE COMMITTEE ON ATOMIC WEIGHTS*

THE Report covers the period from September 30th, 1938, to September 30th, 1939, and the Committee, whose members are the same as before (*cf.* ANALYST, 1939, 64, 352), has been able to complete it, notwithstanding the outbreak of war.

TABLE OF ATOMIC WEIGHTS.—There are three changes in the International Table. The atomic weight of hydrogen is now 1.0080 (previously 1.0081), that of iron 55.85 (previously 55.84), and that of lutecium 174.99 (previously 175.0).

Hydrogen.—The work of different investigators, the most recent of which is that of Swartout and Dole (*J. Amer. Chem. Soc.*, 1939, 61, 2025), on the ratio of the isotopes ^1H and ^2H in certain waters of natural occurrence, has indicated that that value is higher than the one hitherto used for calculating the atomic weight of hydrogen; with the value 1.00785 for ^1H , calculated on the chemical basis, and with the value 6000 for the ratio $^1\text{H}/^2\text{H}$, the atomic weight of hydrogen in natural waters becomes 1.0080.

Iron.—The change in the atomic weight of iron has been based on the work of Hönigschmid and Liang (*Z. anorg. Chem.*, 1939, 241, 361), who prepared spectroscopically-pure metallic iron, converted it into ferrous bromide by heating it in a current of bromine and nitrogen and purified the product by re-sublimation in pure nitrogen. The pure salt, weighed with all precautions, was dissolved in water acidified with sulphuric acid, the solution, which contained no trace of ferric salt, was oxidised with somewhat less than the calculated amount of dichromate, and a nephelometric comparison with silver was made. In 8 experiments the precipitated silver bromide was also weighed. The mean of all the determinations was 55.850, which is slightly higher than the value (55.838) previously found by Baxter, Thorvaldson and Cobb. The Committee attributed this discrepancy to the presence of traces of carbon, and possibly also of ferric salt, in the ferrous bromide used by Baxter *et al.*

Baxter and Hoover (*J. Amer. Chem. Soc.*, 1912, 34, 1857) obtained the value 55.847 by reduction of ferric oxide, and Hönigschmid, Birckenbach and Zeiss (*Ber.*, 1923, 56, 1473), basing their results on the analysis of ferric chloride, found the value to be 55.853.

The most recent investigations of the quantitative ratios of the isotopes of iron have given the values 55.853 and 55.851. The Committee therefore adopted the value 55.85 for the Table of Atomic Weights, since, although the maximum value obtained by the reduction of ferric oxide is 55.847, the possibility of the reduction not being quite complete cannot be excluded.

Lutecium.—The decision to change the atomic weight of lutecium was based on the work of Hönigschmid and Wittner (*Z. anorg. Chem.*, 1939, 240, 284), who prepared pure lutecium trichloride and titrated it nephelometrically; in some of the experiments the precipitated silver chloride was also weighed. The resulting value, 174.996, agrees well with the value obtained spectrographically by Mattauch and Lichtblau (*Z. Physik.*, 1939, 111, 514), 174.994. The Committee therefore adopted the value 174.99 for the Table.

Other Elements.—Among other atomic weight investigations reviewed by the Committee were those on chlorine by Hönigschmid and Wittner (*Z. anorg. Chem.*, 1939, 242, 222), on molybdenum by Mattauch and Lichtblau (*Z. physik. Chem.*, 1939, 42, B, 288), on europium by Lichtblau (*Naturwiss.*, 1939, 27, 260), and on lead by Nier (*Physical Rev.*, 1939, 55, 193). The results did not indicate the necessity for any immediate changes in the respective values in the Table.

Union of South Africa

FERTILISER PROBLEMS IN VEGETABLE PRODUCTION†

QUESTIONS of industrial development and of local markets are surveyed, and an outline is given of overseas practices in the period before the war. From the records of the experience gained the general conclusion is drawn that if vegetable growing is to be profitable, an artificial fertiliser should be applied, if necessary, only as a supplement to, or partial substitute for, natural manure. Green manuring as a method of building up soil organic matter in South Africa is not to be recommended, since the humus is being oxidised too rapidly. Vegetable gardeners near inland towns make use of sewage sludge to a limited extent, but some of the coastal municipalities in South Africa have their sewage pipes running into the sea, so that this source of manure supply is not available in those areas. Composting is being practised on an ever-increasing scale, and gives results as good as those obtained with manure.

pH OF SOILS.—The Virginia Truck Station of U.S.A. had drawn up the following table for

* *J. Chem. Soc.*, 1940, 475.

† Reprint No. 19, 1940. By E. J. Greenstein, Division of Chemical Services, Pretoria.

soils in coastal plains, vegetable crops being grouped according to ranges of soil reaction for optimum growth:

pH 5.0 to 5.5.—Potatoes, sweet potatoes, water melons.

pH 5.5 to 6.5.—Beans, broccoli, cabbage, carrots, cauliflower, cucumber, parsley, parsnips, pumpkins, radishes, sweet corn, squash, tomatoes.

pH 6.0 to 6.5.—Asparagus, beets, celery, leeks, lettuce, onions, peas, spinach.

In South Africa the pH values can be raised somewhat. For example, in certain northern Transvaal areas tomatoes grow well at a pH range of 6.2 to 7.0.

There are definite signs that some of the soils in the Johannesburg and Pretoria districts are very acid, the pH values ranging from 4.5 to 6.5 in the vegetable-producing areas.

GENERAL RECOMMENDATIONS.—The use of acid-forming fertilisers must be avoided on acid soils; conversely, alkaline fertilisers should not be used on limy soils. For acid soils and for vegetable crops, it is generally considered that the fertiliser should contain a relatively large proportion of phosphorus, the ratios between the nitrogen, phosphoric oxide and potassium oxide being somewhat as follows:—4 : 12 : 4; 4 : 16 : 4; 4 : 10 : 6; 3 : 12 : 6. For potatoes and root crops, especially on sandy soil, fertilisers containing a large proportion of potash are preferred (4 : 8 : 8; 5 : 10 : 10).

In general, a heavy dressing of slow organic material, such as manure or compost, plus a good dressing of phosphate and potash at planting time is advocated, together with top dressings of sulphate of ammonia (or nitrate of soda or nitro chalk in very acid soils) when necessary.

COMPOST: ITS PREPARATION AND USE*

This pamphlet describes the nature and object of compost making, and discusses briefly the relative merits of organic manures and inorganic fertilisers.

FATE OF ORGANIC MATTER IN THE SOIL.—The following table shows how the organic matter content (both carbon and nitrogen) of the soil is a fairly reliable guide to its state of fertility, and also demonstrates the rapid loss of organic matter under arable conditions:

| | Carbon Per Cent. | Nitrogen Per Cent. |
|---|---------------------|-----------------------|
| No. 1. Virgin land (veld) | 1.20 | 0.098 |
| No. 2. Abandoned land, previously cultivated | 0.55 | 0.050 |
| No. 3. Land from same slope as No. 2, but still under cultivation | 0.89 | 0.071 |

It is difficult to build up the organic matter of soil by green manuring; normally, at least 75 per cent. of carbon added in that form will be lost by the oxidising action of the soil within 4 months of ploughing in the green crop.

METHODS OF MAKING COMPOST.—The essential conditions for the making of synthetic manure or compost include air supply, suitable temperature, adequate moisture supply, addition of suitable mineral nutrients, maintenance of a suitable reaction in the fermenting heap, and the presence of living micro-organisms. Each of these factors is discussed in detail.

In the ADCO process a patent nutrient mineral mixture is used to induce and maintain decomposition of the plant residues, whilst in the INDORE process a suitable proportion of inoculum, usually containing a good supply of available nitrogen (e.g. from manure or urine earth from the floor of cattle sheds), together with wood ashes, is used. Directions as to the factors necessary for the successful working of each process are given. Modifications suitable for special local conditions or material are also described.

MANURIAL VALUE OF COMPOST.—Well-made compost should have a nitrogen-content closely approximating that of good farmyard manure. The phosphate-content is nearly always low, unless that constituent is specially added, as in the ADCO process. The potash-content also tends to be low, although that is not so important as the nitrogen and phosphate contents in South Africa, where the soils contain a fair proportion of potash. One of the main factors influencing the final composition is the amount of soil that has been allowed to mix with the compost. In the INDORE system, soil may amount to 50 per cent. of the dry weight of the compost. The lowering of nitrogen, phosphate and potash contents, as revealed by a chemical analysis, will not necessarily reflect the efficiency of the composting process unless total weights are supplied, so that a balance sheet may be made out. The presence of an unduly high proportion of soil will naturally increase the cost of carting and handling.

The following Table shows the composition of various types of compost and manure:

* Science Bull. No. 201. Department of Agriculture and Forestry (*Chemistry Series No. 157*). By E. R. Orchard, Ph.D., Division of Chemical Services, Pretoria.

| Description | Composition on air-dry basis | | | | |
|---|------------------------------|----------------|-----------|-------------------------------|------------------|
| | Moisture | Organic matter | N | Available | |
| | | | | P ₂ O ₅ | K ₂ O |
| | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. |
| Well conserved cattle manure | 80.5 | 78.5 | 2.2 | 1.0 | 2.2 |
| ADCO from wheat straw (England) | 75.0 | 56.0 | 2.4 | 1.7 | 0.8 |
| ADCO from grass cuttings (England) | 82.7 | 58.4 | 4.9 | 3.6 | 5.9 |
| ADCO from garden refuse (England) | 63.7 | 44.9 | 1.5 | 2.6 | 3.4 |
| ADCO from wheat straw (South Africa) | 79.6 | 69.5 | 2.4 | 1.7 | 3.1 |
| ADCO from fresh veld grass (Rhodesia) | 72.9 | 57.1 | 2.0 | 1.7 | 1.1 |
| ADCO from maize stalks (Rhodesia) | 72.4 | 58.9 | 2.2 | 1.3 | 1.0 |
| ADCO from sugar-cane trash (South Africa) | 62.4 | 56.7 | 1.9 | 1.6 | 1.4 |
| Kraal refuse (South Africa) | — | 48.4 | 1.5 | 0.2 | 1.8 |
| Farm wastes put through kraal | — | 29.4 | 0.8 | 0.2 | 1.3 |
| INDORE compost from maize and sunflower stalks | 60.8 | 39.5 | 1.1 | 0.2 | 0.7 |
| INDORE compost, mean of 12 samples made largely from kraal wastes | — | 32.5 | 0.9 | 0.1 | 0.5 |
| Guano, cull oranges, veld grass and pine needles | — | 37.5 | 1.6 | 1.4 | 1.2 |

The variable nature of compost and its low phosphate-content used are clearly brought out. The age of the vegetable residues has a pronounced influence on composition.

Over-mature compost closely resembles a garden soil and will not repay its cost of manufacture. Various chemical methods of judging the state of maturity have been suggested, but these are not of practical value on the farm. Usually compost is regarded as suitable for carting when 80 to 90 per cent. is fine enough to pass through a $\frac{1}{4}$ -inch mesh and when the coarsest constituents have rotted so that they can be pulverised easily by rubbing.

Steps must be taken to minimise the loss, by excessively rapid or unnecessary oxidation, of organic matter both from the soil and from the compost heap.

The pamphlet concludes with sections on crop response to compost applications and on composting from the economic aspect, and a bibliography of 19 references.

Norwegian Canning Industry

SIGNIFICANCE OF THE BACTERIAL COUNT IN DETERMINING THE FRESHNESS OF HERRING AND BRISLING, THE RAW MATERIALS OF KIPPERED HERRINGS AND CANNED BRISLING*

BACTERIAL DECOMPOSITION OF WINTER HERRING.—To establish a standard for the quality of fresh winter herring, the raw material of kippered herrings, the fish were stored at temperatures ranging from -3° to $+13^{\circ}$ C., and samples were taken daily for bacterial count, organoleptic examination and smoking. Bacterial invasion of the flesh was investigated by aseptically taking portions of the back after removal of the skin. The fish, clipped with sterile scissors, were shaken with sterile saline and plating out was done on agar and gelatin. The results of the bacterial counts, which are given in tables, show good correlation with the degree of freshness. When the fish give a count of 500,000 per g. their flavour is "strong"; stale fish contain 1 to 2 million bacteria per g. and may be considered unmarketable. The examination of a mixed sample of 6 lots of fish from different factories showed that 1000 to 100,000 bacteria per g. is the average number for herring of good quality. The bacterial count of smoked herring clearly shows the bactericidal efficacy of the smoking, the average count being about 1000 per g.

SANITARY QUALITY OF BRISLING USED IN THE NORWEGIAN CANNING INDUSTRY.—Brisling, which are packed during the summer, are very subject to invasions of decomposing bacteria, and precautions are taken to guard against this during transport. Samples from four factories were examined, an average of ten fish being used for each sample. The whole fish, including intestines and surface slime, were clipped with sterile scissors and shaken with sterile saline, and dilutions were made for plating; the counts are therefore not comparable with those obtained with herring, and the average for fish of good quality one day old was found to be about 100,000 per g.

* Bull. No. 57, Norwegian Canning Industry. E. Aschehoug and R. Vesterhus. *Tidsskrift for Hermetikindustri*, 1940, 17-26; 49-53.

PROCESSING STUDIES OF KIPPERED HERRING.—It was found that "high-short" processing impaired the quality of the smoked fish. Processing at 6 lbs. pressure for 70 minutes in half-oval tins is recommended. For smoked brisling packed in tomato sauce, processing at even lower temperature for a longer time is recommended (e.g. at 4 to 6 lbs. pressure for 60 minutes), the colour and taste of the tomato sauce being easily impaired; but for packing in olive oil, processing at 6 to 8 lbs. pressure for 50 minutes is permissible. D. R. W.

British Standards Institution

THE following Standard Specification has been issued*

No. 894—1940. THE DETERMINATION OF THE FLOW AND DROP POINTS OF FATS AND ALLIED SUBSTANCES

(APPARATUS AND METHOD OF USE)

The apparatus, which is of the Ubbelohde type, is shown in diagrams. A glass cup of specified dimensions, with an orifice at the bottom, is filled with the fat to be tested and attached to a metal fitting which is fixed to a standard thermometer, the bulb of which is situated centrally in the cup. The thermometer, with the cup attached, is fitted through the cork of a boiling tube at a specified level, and the boiling tube is fixed vertically in a beaker containing a suitable liquid heating medium which is kept stirred. The temperature of the outer bath is adjusted so that the temperature of the fat rises at the rate of 1° C. per minute over a range of about 10°C. immediately below the flow-point.

The *flow-point* is defined as the temperature at which the substance under examination forms an approximately hemispherical protuberance at the orifice of the glass cup. The *drop-point* is the temperature at which the first drop falls from the glass cup.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Detection by Walkiewicz's Method of the Decomposition of Meat. H. Keller and H. Möller. (*Z. Fleisch- u. Milch-hyg.*, 1939, 49, 141-143; *Z. Unters. Lebensm.*, 1940, 79, 298-299.)—Walkiewicz's method, which consists in adding 2 or 3 drops of extract from the meat to 3 or 4 ml. of mercuric chloride solution (1 : 1000), and to the same volume of the mercuric solution containing acetic acid (0.05 : 1000), is modified by using an extract prepared by shaking 5 g. of meat with 50 ml. of water. At pH values not lower than 6.2 there forms immediately a grey-violet precipitate, which settles in a few seconds and, on stirring, yields an opalescence. With acidified sublimate solution this reaction is positive only with meat in which the acid-forming stage of decomposition is past. The authors' results, which do not agree entirely with previous work, are as follows:—(1) Muscular tissue containing much connective tissue, fat and blood always gives a positive reaction with both sublimate solutions, so that decomposition of such meat cannot be detected by this method. (2) With beef and calf muscle from healthy animals with a normal meat acidity Walkiewicz's reaction detects decomposition 1 or 2 days earlier than the Eber (hydrogen-sulphide) test. The pH at which the precipitate forms is 6.0. (3) Beef and veal from unhealthy animals give a positive reaction with both sublimate solutions, indicating very slight decomposition. (4) Pig muscle and slightly acid horse muscle, from both healthy and unhealthy animals, give a positive reaction with both sublimate solutions, so that the test is not applicable to pork or horseflesh. E. M. P.

Estimation of Sulphur Compounds in Vegetables. W. Diemair and J. Koch. (*Z. anal. Chem.*, 1940, 119, 94-108.)—Conversion of the loosely combined sulphur in the proteins of vegetable substances into thiosulphate by treatment with alkali, with subsequent transformation into ferric thiocyanate, was found to be an unsuitable method for colorimetric estimation. The following modification of the method of Diemair, Strohecker and Keller (*Z. anal. Chem.*, 1939, 116, 385) proved more satisfactory. The material (20 g.) is mixed with 50 ml. of sodium hydroxide solution (0.1 N or 0.01 N) and distilled in a pear-shaped 500-ml. flask over a naked flame at such a rate that distillation begins in 4 minutes and 18 ml. of distillate are collected in the next 6 minutes. The distillate is received in a cylinder containing 1 ml. of a 0.5 per cent. solution of dimethyl-*p*-phenylenediamine in conc. hydrochloric acid and 1 ml. of 10 per cent. ferric chloride solution.

* Obtainable from the Publications Department, 28, Victoria Street, London, S.W.1. Price 2s. net; post free 2s. 2d.

The methylene blue formed is estimated colorimetrically in a Pulfrich photometer. Strict adherence to the experimental conditions is necessary, especially to the ratio of material to alkali, the time of distillation and the volume of distillate. The sample must be so prepared that it forms a thin broth with the alkali. As it was found that, of the vegetables investigated, peas produced much frothing, the quantity of sample and alkali used were reduced to 10 g. and 25 ml. respectively, and the vegetable was not pulped before distillation. With canned vegetables the maximum separation of sulphur occurred when the alkali solution was 0.01 *N*, but with fresh vegetables 0.1 *N* alkali was required. A number of fresh and preserved vegetables were examined by the process described, and the amount of loosely combined sulphur found was compared with the total sulphur determined by the method of Denis (*J. Biol. Chem.*, 1910, 8, 401). With spinach, asparagus, carrot, pea, mushroom, bean, kohlrabi and celery the relation between the loosely combined sulphur and the total sulphur was not the same, but the experimental results indicated a division of these vegetables into three groups. In the first group (asparagus, pea and kohlrabi) the loosely combined sulphur is greater in the conserve than in the fresh vegetable, and in the second group (spinach and bean) there is less in the conserve than in the fresh substance. In the third group (carrot, mushroom and celery) no sulphur was obtained by distillation. This grouping can be explained by a consideration of the processes of manufacture of the conserves. Examination of peas in different stages of ripeness showed that, although the total sulphur increased with advancing ripeness, the sulphur obtained by distillation diminished. In order to determine the relation of the loosely combined sulphur to the cystine, cysteine and glutathione in the plant, the proteins of spinach, asparagus and pea were hydrolysed and the cystine and cysteine were separated by means of 9-phosphotungstic acid and estimated colorimetrically by the methods of Schöberl and Ludwig (*Ber.*, 1937, 70, 1422) and Hornung (Dissertation, Würzburg, 1938). The ratio of sulphur derived from cystine to that derived from cysteine, although subject to variation, was found to approach 1 to 6 in the vegetables investigated. In asparagus and peas the sulphur was entirely in the organic form. In beans 85.9 per cent., in kohlrabi 81.3 per cent., in carrot 79.3 per cent. and in spinach 63.8 per cent. of the sulphur was in the organic form. These results suggest that in those portions of the plant where there is meristematic tissue and active growth (asparagus tips) or nutritional material for the embryo (pea) the sulphur is in the easily assimilated organic form. In leaf (spinach), root (carrot) and stem (kohlrabi), where growth is not active, a smaller proportion of the sulphur is in the organic form. A. O. J.

Component Glycerides of Vegetable Fatty Oils. Niger-seed Oil. N. L. Vidyarthi and M. V. Mallya. (*J. Indian Chem. Soc.*, 1940, 17, 87-95.)—The component fatty acids are estimated by the usual methods of lead salt separation, ester fractionation and hexabromide determination. The neutral oil is then dissolved in 6 times its weight of dry acetone and crystallised at 0° C., when any fully saturated or di-saturated glycerides will crystallise. Successive oxidation of the neutralised oil in acetone solution affords a measure of the quantity of fully-saturated glycerides. The remaining oil, after removal of the saturated and disaturated glycerides, is dissolved in light petroleum spirit and brominated at 0° C., and the solid and liquid bromoglycerides are further resolved into a number of fractions by treatment with benzene, acetone, alcohol and acetone-alcohol (1:1 mixture). The fractions are all brominated, the component fatty acids are determined, and the amounts of the different glycerides in the fractions are calculated. This method has been used for niger-seed oil (*Guizotia abyssinica*) which was extracted from the seed with carbon tetrachloride. The sample examined had the following characteristics: n_D^{25} , 1.472; saponification value, 189.7; iodine value, 129.2; acetyl value, 19.8; free fatty acids (oleic), 4.27. The percentages of glycerides in the oil are given as tri-linolin, 2; myristo-di-linolin, 2; myristo-oleo-linolin, 3; palmito-di-linolin, 6; stearo-di-linolin, 2; palmito-oleo-linolin, 11; stearo-oleo-linolin, 4; di-oleo-linolin, 30; and oleo-di-linolin, 40. Palmito-glycerides contain the small quantity of lauric, and other lower acids present, and stearo-glycerides contain the arachidic, lignoceric and behenic acids. By hydrogenating the oil to iodine values of 1.8 and 27.2 respectively the amount of C_{18} glycerides was calculated to be about 77.3 per cent. and the remaining 20.8 per cent. of glycerides to contain at least one acid with less than 18 carbon atoms. This agrees fairly well with results obtained from the brominated glycerides. The major component acids—oleic and linolic—are evenly distributed in the glycerides. D. G. H.

Carbohydrate Characterisation. I. Oxidation of Aldoses by Hypiodite in Methanol
II. Identification of Seven Aldo-Monosaccharides as Benzimidazole Derivatives.
 S. Moore and K. P. Link. (*J. Biol. Chem.*, 1940, 133, 293-311.)—A very satisfactory method of identifying aldoses consists in oxidation to the corresponding acids by means of potassium hypiodite and methylalcohol, and condensation of the acid so obtained with *o*-phenylenediamine to give the benzimidazole derivative. I.—A sample of the solution or syrup equivalent to 2 g. of aldo-hexose is concentrated or diluted, as required, to a volume of 4 ml. Into a 500-ml. 3-necked flask equipped with a stirrer, thermometer and dropping-funnel are introduced 5.7 g. of iodine and 80 ml. of methyl alcohol. After being stirred for a few minutes the solution is warmed on the water-bath to about 40° C., and the aldo-hexose solution, dissolved in 25 ml. of methyl alcohol (filtered and decolorised with charcoal if necessary), is added. Stirring is at once resumed, and 4 per cent. potassium hydroxide solution is added dropwise, 65 ml. over a period of 10 to 15 minutes, and, after 10 minutes' further stirring, 50 ml. more, also dropwise; the colour

should then be pale straw yellow, and if it is darker a few more ml. of the alkali may be added. Finally the liquid is stirred for 10 minutes and allowed to cool, and any precipitate that separates is filtered off, washed twice with methyl alcohol and once with water, and dried. The solid consists of the potassium salts of the acids derived from glucose, galactose and arabinose. The filtrate is returned to the reaction flask, and a solution of 5 g. of barium iodide (dihydrate) in 25 ml. of methyl alcohol is added to it dropwise with stirring. The barium salts corresponding to any other aldoses present (except xylose) separate out and are centrifuged off and washed with methyl alcohol and ether.

II.—The mixed potassium or barium salts are condensed with *o*-phenylenediamine in presence of acid catalysts at 135° C., except the salt of xylonic acid, which requires zinc chloride and a temperature of 180° C. The benzimidazoles crystallise readily, have sharp melting-points, and yield derivatives that crystallise well. They possess many advantages over osazones for characterising sugars. Moreover, benzimidazoles can be precipitated as copper salts from aqueous solution, thereby facilitating the isolation of small amounts of derivatives which might otherwise escape detection; the benzimidazole is regenerated from the copper salt by the use of hydrogen sulphide. The following is a summary of the properties of the benzimidazole derivatives from seven aldomonosaccharides:

| Carbohydrate | Benzimidazole | | Hydrochloride | Picrate |
|---------------------|---------------|-------------------|---------------|---------------|
| | m.p. °C. | $[\alpha]_D^{25}$ | m.p. °C. | m.p. °C. |
| <i>l</i> -Arabinose | 235 (decomp.) | + 49.2 | 230 | 158 |
| <i>d</i> -Galactose | 245 (decomp.) | + 43.3 | 202–204 | 217 (decomp.) |
| <i>d</i> -Glucose | 215 | + 9.6 | 180 | 203 (decomp.) |
| <i>d</i> -Lyxose | 189 | – 12.8 | 191 | 95–99 |
| <i>d</i> -Mannose | 227 (decomp.) | – 22.0 | 101–150 | 205 (decomp.) |
| <i>l</i> -Rhamnose | 207 | + 27.4 | 173–174 | 168 |
| <i>d</i> -Xylose | 224 | + 64.8 | 200–202 | 191 |

The rotations were measured in 5 per cent. citric acid solution at $C = 2$. The method of carrying out the condensation is best illustrated by the use of calcium gluconate. To 2 g. of calcium gluconate (hydrate), (0.009 mole of gluconic acid), in a test-tube, 1.1 g. (0.01 mole) of *o*-phenylenediamine, 4 ml. of water, 1 ml. of ethyl alcohol, and 1.7 ml. (0.02 mole) of conc. hydrochloric acid are added. The mixture is heated for 2 hours in an oil-bath at 135° ± 5° C. The syrup that remains is dissolved in 10 ml. of water, charcoal is added, and the suspension is filtered. On making the filtrate (diluted to about 30 ml.) alkaline with ammonium hydroxide, crystals of gluco-benzimidazole separate out. The treatment of the potassium salt fraction from the oxidation of aldoses is carried out in a similar manner, but with the addition of 0.8 ml. of syrupy phosphoric acid. The working up of the barium salt fraction is carried out in a slightly different manner. The equivalent of 1 g. of aldo-hexose is suspended in 10 ml. of water in a centrifuge tube, and the solution is neutralised to phenolphthalein with dilute hydrochloric acid. To the neutral solution 0.8 ml. of conc. hydrochloric acid is added and sufficient of a mixture of sulphuric acid and water (1:1) to precipitate the barium ion. The solution is filtered, the filtrate is evaporated to 4 ml., and the concentrate is transferred to a test-tube containing 0.7 g. of *o*-phenylenediamine. After the addition of 0.5 ml. of phosphoric acid the condensation is continued as described above. To the filtrate from the potassium or barium salt fraction, from which excess ammonia has been removed by evaporation, a cupric ammonia solution is added, 20 ml. for each g. of benzimidazole present. This solution is prepared by suspending 10 g. of cupric acetate (monohydrate) in water, adding sufficient ammonia to give a clear solution, and diluting to 100 ml. The precipitate of copper salt is filtered off and suspended in a mixture (3:1) of water and alcohol. The suspension is decomposed with hydrogen sulphide, and the filtrate usually crystallises on being concentrated. Xylonic acid, if present, does not yield a benzimidazole under these conditions, but an intermediate reaction product is formed which is contained in the filtrate obtained by decomposing the copper salt with hydrogen sulphide. Accordingly, the residue left after the removal of any benzimidazoles is concentrated to a syrup, and for each 0.5 g. of xylose estimated to be present, 0.5 ml. of conc. hydrochloric acid and 0.3 g. of zinc chloride are added. The mixture is placed in an oil-bath at 135° C., and the temperature is raised to 180° C. over a period of 45 minutes and maintained at that point for an hour. The syrup is taken up in water and decolorised. Ammonia is added to the solution and excess removed by evaporation. The precipitate of zinc benzimidazole and zinc hydroxide is filtered off, suspended in water and decomposed with hydrogen sulphide. The filtrate yields crystals of xylo-benzimidazole on concentration. The presence of fructose does not interfere with the isolation of aldo-benzimidazoles, but a small amount of *d*-arabo-benzimidazole (m.p. 235–236° C., $[\alpha]_D^{25} = -51^\circ$; hydrochloride, m.p. 229° C.; picrate, m.p. 158° C.) is obtained from the barium salt fraction. Care should therefore be exercised in drawing conclusions from the formation of this compound, and tests for pentoses and ketoses should be carried out on the original material to determine whether fructose or *d*-arabinose is the source of this derivative.

F. A. R.

Behaviour of Olive Oil and other Oils with Antimony Trichloride. W. H. Dickhart. (*Amer. J. Pharm.*, 1940, 112, 131-133.)—Antimony trichloride produces a blue colour with vitamin A, synthetic vitamin A, and various carotenoids and other sterols. A standardised procedure was applied to a large number of olive and other oils. A 33.33 per cent. solution of antimony trichloride was made with C.P. chloroform, and to 1 ml. of this solution 2 g. (approx. 50 drops) of oil were added, the mixture was shaken and left for 1 hour, and the colour reading was taken in a Lovibond tintometer. Of 9 virgin olive oils from various sources, 7 gave an emerald-green colour, one ("Greek, extra") a pea-green, and one (from California) a blue-green colour. Five samples of refined olive oils (Greek, Spanish, Italian, Tunisian and Californian) gave blue colours, and one extracted refined foots oil an olive-green colour. The unsaponifiable matter from virgin oil gave a red colour changing to blue, and that from refined olive oil a red colour. A refined olive oil giving a blue colour was heated to 220° C. for 20 minutes, and then gave an amber colour. It is suggested that olive oil contains one or more of the lipochromes, as they are destroyed by oxidation, reduction, high temperatures and excessive exposure to light; and thus olive oil would presumably contain vitamin A, or a substance converted into vitamin A by the liver. The green colour is regarded as probably due to β -carotene or a mixture of a yellow pigment with the blue colour from α -carotene, γ -carotene or squalene. A list is given of the colours produced with a large number of oils, normal and hydrogenated, and it is suggested that the method may be of value in distinguishing normal olive oil from teased oil (orange with fluorescence); hydrogenated fish oil (deep purple) from hydrogenated cottonseed oil (light pink); oiticica and China wood oils (red) from those of lumbang (yellow to dark red), perilla (yellow to pale pea-green) and linseed (brown); cottonseed oil (pink) from kapok oil (yellow). D. G. H.

Fatty Oil from the Seeds of *Bauhinia variegata*. S. V. Puntambeka and S. Krishna. (*J. Indian Chem. Soc.*, 1940, 17, 96-100.)—*Bauhinia variegata*, Linn. (N.O. Leguminosae) is a deciduous tree found in the sub-Himalayan tract from the Indus eastward, and known in Hindi as Kachnar; its flowers are eaten as a vegetable. It produces pods, 6-12 inches by $\frac{1}{2}$ to 1 inch, containing 10 to 15 seeds. The seeds consist of 20 per cent. endocarp and 80 per cent. of kernels, which yield about 16.5 per cent. of a pale fatty oil on extraction with petroleum spirit, and about 6 per cent. when expressed. The oil examined had the following characteristics:—sp.gr. at 30°C., 0.9206; n_D^{20} , 1.4603; iodine value (Hanus), 91.3; Hehner value, 92.0; acid value, 2.8; unsaponifiable matter, 1.6 per cent. The mixed acids had mean molecular equiv. 294 and iodine value (Hanus) 93.2; they consisted of 32.3 per cent. of saturated and 67.7 per cent. of unsaturated acids. The mixed fatty acids were separated by the lead salt method, and the methyl esters of the solid acids were distilled into 5 fractions, which were analysed. The liquid acids were oxidised in cold alkaline solution with potassium permanganate, and the freshly prepared mixed acids were also brominated. The unsaponifiable matter appeared to contain sitosterol, and the residue was probably mostly hydrocarbons. The proportions of the constituent acids are given as: myristic, 1; palmitic, 17; stearic, 13.4; lignoceric, 1; oleic, 31.8; linolenic, 35.9 per cent. D. G. H.

Oil from the Fruit of *Ferula alliacea*. P. K. Bose and S. N. Dutt. (*J. Indian Chem. Soc.*, 1940, 17, 49-52.)—The mature fruit of the umbelliferous plant, *Ferula alliacea*, Biss, has a pronounced aromatic odour when crushed. A petroleum spirit extract yields on steam distillation about 0.9 per cent. of a colourless sweet-smelling volatile oil. The fruit, which also contains some natural coumarins, yielded on extraction with petroleum spirit 19 per cent. of a yellow, bitter-tasting oil with the following constants:—sp.gr. at 31.5/31.5°C., 0.9156; n_D^{20} , 1.4691; solubility in alcohol at 25°C., 2.55 per cent.; saponification value, 189.62; iodine value (Wijs), 90.73; acetyl value, 23.56; Reichert value, 1.81; Polenske value, 0.25; acid value, 16.6; unsaponifiable matter, 1.96 per cent.; bromide test, nil; total saturated acids (corr.) 14.02 per cent. with iodine value, 19.9; unsaturated acids, 78.08 per cent. with iodine value, 100.6. The solid saturated acids contained some unsaturated acids, such as erucic or petroselinic acid, the lead salts of which are insoluble in ether. Five fractions were obtained after alcoholysis. The first consisted mainly of essential oil. Myristic and palmitic acids were apparently absent, and fraction V (b.p. 210-220°C.), which constituted the bulk of the distillate, had iodine value 86.16, and its separated fatty acids did not yield a solid product on bromination. D. G. H.

Quantitative Determination of Powdered Cinnamon and Cassia. A. H. Saber. (*Quart. J. Pharm.*, 1940, 13, 7-13.)—The phloem fibres of cinnamon and cassia bark occur isolated or in rows of single files. From the areas of fibres per g. of the powdered bark, these drugs may be quantitatively determined in powders (*cf.* Saber, *Quart. J. Pharm.*, 1934, 7, 645; *Abst.*, *ANALYST*, 1935, 60, 258). Twenty g. of cinnamon quills were reduced to No. 60 powder, 5 g. of this were reduced to No. 85 powder and dried at 100°C., and 0.1 g. of the dried powder was thoroughly mixed with 0.05 g. of lycopodium. The mixture was cleared in a small glass tube with 3 to 3.5 ml. of chloral hydrate solution (5 : 2), and the preparation was made up to 10 ml. with a suspending liquid (2 vols. of glycerin, 1 vol. of tragacanth mucilage and 2 vols. of water). From this suspension mounts were made in which the areas of fibres were determined (for details see Wallis and Saber, *Quart. J. Pharm.*, 1933, 6, 655, or B.P. Codex, 1934, Appendix IX, 1592). The

minimum area representative of each mount was about 30 sq. mm. (actually 33-152 sq. mm.), consisting of 7 strips across the counting square separated by 2-mm. intervals. Commercial cinnamons of various qualities and grades were similarly examined. Mean results for 6 samples were: Good quality commercial (A) 92.5, Grade 0000 (B) 98.0, Grade 00 (C) 87.0, Quillings (D) 110.0, Featherlings 70.0, Chips 40.0 sq. cm. per g. The high result for (D) was due to the presence of an unknown bark. When unground, the ungraded sample (A) was less fine than (B) but better than (C). The relation between the results and the qualities and grades of the powders is due to the presence of the outer bark, which is fibreless, in increasing amounts as the quality of the sample deteriorates. It is not necessary to remove the small amount of oil in the bark before examination. Cassia bark or Chinese cinnamon is the partly decorticated dried bark of *Cinnamomum Cassia* Blume, and is very closely related to *Cinnamomum zeylanicum*, from which cinnamon is obtained. When examined by the method above, the average result for 2 cassia samples was 13.1 sq. cm. per g.; this is so different from that of good quality cinnamon—e.g. 92.5 on 3 preparations of (A)—that the relative amounts of cassia and cinnamon in a mixture of both may be determined by this count. In an experimental cassia-cinnamon mixture the cassia found by this calculation was 33.4 per cent. (actual, 35.38 per cent.). When the grade of cinnamon is not known the calculation should be based on results for quills, because lower grades are not permitted in medicine. If the cassia is powdered by a disintegrator instead of an iron hand-mortar, results are higher—e.g. 15 sq. cm. per g. E. B. D.

Determination of Piperazine. A. Castiglioni. (*Z. anal. Chem.*, 1940, 119, 118-120.)—Pratt and Young (*J. Amer. Chem. Soc.*, 1918, 40, 1428) have shown that piperazine yields a characteristic crystalline precipitate with Dragendorff's reagent, and other authors have suggested that this extremely insoluble substance might be used for quantitative purposes. It was found, however, that under ordinary conditions, the piperazine is not completely precipitated. Good results have been obtained by the use of the reaction of piperazine with carbon disulphide in which the addition compound, $C_4H_{10}N_2CS_2$, is formed. The precipitation must be made with an excess of carbon disulphide in absence of water. In the application of the reaction to the determination of carbon disulphide (Castiglioni, *Z. anal. Chem.*, 1939, 115, 257; Abst., ANALYST, 1939, 64, 230) 95 per cent. alcohol or mixtures of other solvents may be used, but in the determination of piperazine only chloroform or mixtures of other solvents (alcohol-ether, alcohol-acetone) give satisfactory results, the reason being the limited solubility of carbon disulphide in alcohol containing water. Chloroform is recommended as solvent because it promotes the formation of a coarse precipitate, which settles rapidly. The procedure is as follows:—If the piperazine is dissolved in 95 per cent. alcohol, an excess of a mixture of equal parts of carbon disulphide and ether is added and the mixture is warmed slightly and allowed to stand. If the piperazine is in chloroform solution it is sufficient to add an excess of carbon disulphide, warm slightly and allow the mixture to stand. When the supernatant liquid has become clear, the precipitate is collected, washed with small amounts of alcohol-ether or chloroform (according to the solvent used), dried at 105° C. and weighed. Each molecule of the addition compound contains a molecule of piperazine. Hexamethylene tetramine gives no precipitate with carbon disulphide; consequently, the method may be used for the determination of piperazine in the presence of that substance. A. O. J.

New Solanaceous Alkaloids from *Duboisia myoporoides*. W. Mitchell. (*Pharm. J.*, 1940, 144, 137.)—As previously reported (*J. Chem. Soc.*, 1937, 1820; 1938, 1685), the alkaloids in this drug are hyoscyne and the four new alkaloids: tigloidine, valeroidine, poroidine and isoporoidine, but no trace of hyoscyamine or other similar alkaloid reported by earlier workers was found. It is therefore recommended that the official description of "duboisine" sulphate as a mixture of hyoscyne and hyoscyamine sulphates be discontinued. Tigloidine has now been synthesised and shown to be tiglyl- ψ -tropine; valeroidine is the monoisovaleryl ester of a dihydroxytropine previously isolated from Peruvian coca leaves as the dibenzoyl ester, and has also been synthesised. Poroidine and isoporoidine have been isolated as a mixture originally called base Z; the former is now shown to be isovalerylnortropine, and the latter d - α -methylbutyrylnortropine, and both have been synthesised. A partial separation of base Z, which closely resembles a mixture of 10 parts of poroidine and 1 part of isoporoidine, has been made by an indirect method, and isovalerylnortropine has been isolated. The Codex indicates that duboisine is a mydriatic drug and much more powerful than atropine. D. G. H.

Pelletierine of Commerce. J. A. Goodson. (*Quart. J. Pharm.*, 1940, 13, 57-63.)—In the B.P., 1932, pelletierine means the total alkaloids of the pomegranate stem and root bark; the French Codex drug is a more basic fraction of the total alkaloids of the bark. In this article pelletierine means Tanret's pure, optically active base, isolated from pomegranate root bark, and "pelletierine" is used for the alkaloids liberated from pomegranate root bark by sodium hydroxide but not by sodium bicarbonate. The weaker bases of the alkaloids (pseudo-pelletierine and methyl pelletierine) are probably valueless as vermicides, and the anthelmintic properties of the bark appear to be due to the presence of l -pelletierine and its racemic isomer. It is therefore considered desirable that some quantitative test, such as the determination of the bases liberated by sodium hydroxide but not by sodium bicarbonate, should be introduced into the B.P., especially

as commercial sulphates and tannates recently examined have frequently been deficient in "pelletierine." Ewers' method gives fairly accurate results. The inactive pseudo-pelletierine may be separated from the total alkaloids by treatment of the hydrochlorides with acetone, in which pseudo-pelletierine hydrochloride is insoluble. F. B. D.

Volumetric Determination of Acridines with Methylene Blue. A. Bolliger. (*Quart. J. Pharm.*, 1940, 13, 1-6).—2:8-Diamino-acridine (A) and 2:8-diamino-10-methyl-acridinium chloride (B) and their commercial forms (proflavine, euflavine and acriflavine) are determined by precipitation as picrates and subsequent determination of the excess of picric acid with methylene blue. A solution of 0.1 g. in 30 ml. of 0.5 per cent. acetic acid is precipitated with a measured excess of *N*/100 (2.29 g. per litre) picric acid, diluted to 200 ml., and filtered after precipitation is complete (i.e. after at least 1 hour in a refrigerator). Twenty ml. of the solution are then titrated with *N*/1000 methylene blue by Bolliger's method (*cf. ANALYST*, 1939, 64, 416). The determinations of (A) sulphate, of (B), and of mixtures of the hydrochlorides of (A) and (B) are as described above, except that acetic acid is replaced by water as solvent and that at least 4 hours in the refrigerator are required for complete precipitation. (A), (A) sulphate and (B) are equivalent to 47.85, 32.56 and 38.46 ml. of *N*/100 picric acid solution, respectively, per 0.1 g. The methylene blue solution is standardised on *N*/1000 picric acid solution prepared by dilution of the *N*/100 acid (*cf. Bolliger, loc. cit.*). The monopicrates of (A) and (B) have been isolated and analysed. (A) picrate is obtained as a yellow, partly crystalline precipitate, insoluble in dilute picric acid solution, sparingly soluble in water, alcohol, ether, benzene and chloroform, readily soluble in pyridine; it crystallises from a pyridine-water mixture (1 : 3) in yellowish-orange needles which decompose, without melting, at about 250° C. Its solubility in water at room temperature is less than 0.2 mg. per 100 ml. but if it is recrystallised from hot water its solution remains supersaturated (0.5 to 0.6 mg. per 100 ml.) after standing overnight at room temperature. (B) is obtained as an amorphous orange precipitate, insoluble in dilute picric acid, sparingly soluble in most solvents, soluble in pyridine; it crystallises from dilute pyridine in deep orange-red needles, m.p. 244° C. (with decomposition). Admixture with small amounts (1 : 9) of (A) picrate depresses the m.p. by at least 20° C. Its solubility in water is less than 0.3 mg. per 100 ml. E. B. D.

Bleach Ointment. G. H. Macmorran. (*Pharm. J.*, 1940, 144, 213-214.)—The A.R.P. Handbook No. 3 prescribes a mixture of equal parts by weight of supertropical bleach and white soft paraffin for the preparation of bleach ointment. According to the B.P. Codex, tropical bleach is ordinary bleach mixed with unslaked lime. Two ointments were prepared on a small scale, and their available chlorine contents were determined over a period, with the following results:

| Time Weeks | Tropical bleach ointment | | B.P. bleach ointment | |
|---------------|---------------------------------|--------------------------------|---------------------------------|--------------------------------|
| | Available chlorine Per Cent. | Percentage loss in chlorine | Available chlorine Per Cent. | Percentage loss in chlorine |
| 0 | 16.72 | — | 19.21 | — |
| 2 | 15.49 | 7.36 | 17.31 | 9.89 |
| 4 | 15.36 | 8.13 | 16.47 | 14.27 |
| 6 | 15.09 | 9.75 | 15.65 | 18.53 |
| 8 | 14.73 | 11.90 | 15.10 | 21.40 |
| 10 | 14.50 | 13.28 | 14.57 | 24.15 |
| 12 | 14.31 | 14.41 | 14.12 | 26.50 |

Eight lb. of each ointment were then made in a granite end runner mill, the bleach being passed through a No. 40 sieve before mixing. No difference was observed during milling, the temperature of each ointment remaining about 20° C. Both ointments were left overnight in earthenware jars on a stone floor. In the morning the B.P. bleach ointment had overflowed as the result of vigorous chemical reaction and a "witness-tube" containing phenazone placed in the ointment had melted, showing that the temperature had reached at least 111° C. The ointment had become yellowish and contained only 1.05 per cent. of available chlorine; the tropical bleach ointment contained 16.8 per cent. The effect of the degree of saturation of the base was indicated by the following experiments:—An ointment was made with a vegetable fat having an iodine value of 63, and chlorinated lime containing 39.1 per cent. of available chlorine. At first the chlorine-content was 19.36, but after 2 weeks it had fallen to 11.40 per cent., showing a loss of 41.12 per cent. of the available chlorine. The B.P. ointment in the preceding table, made with chlorinated lime (available chlorine, 39.2 per cent.), was prepared with soft paraffin (iodine value, 5). This ointment lost 9.89 per cent. of its available chlorine in 2 weeks. A third ointment made with hydrogenated fat (iodine value, 1.6) and chlorinated lime (39.1 per cent. of available chlorine) contained 19.04 per cent. of available chlorine, and after 2 weeks showed a loss of only 1.21 per cent. These results confirm those of Fishburn (*Pharm. J.*, 1940, 35), who found that the higher the iodine value of the soft paraffin used, the more rapid the decrease in the available chlorine content. The general conclusions drawn from the experiments are that the use of supertropical bleach is to be recommended, that a soft white paraffin of low iodine value should be used, but that consideration should be given to the possible use of hydrogenated fats.

A tube is the most suitable container for bleach ointment. In the author's opinion the figure of 15 per cent. of available chlorine suggested (see Moir, *ANALYST*, 1940, 154) is too high, and a more reasonable figure would be 10 or 12 per cent.

Tests for Tannic Acid. D. B. Dott. (*Pharm. J.*, 1940, 144, 137.)—Samples of tannic acid which have passed the B.P. test may still be unsatisfactory in practice. Four samples were dissolved in collodion (0.5 g. in 10 ml.). Sample A gave a very turbid solution; B a nearly clear solution in which a deposit slowly formed; C a clear straw-coloured solution; D a clear solution of the usual brownish tint which soon changed to green, although there was no evidence of metallic contamination. Tannic acid should dissolve readily in acetone giving a syrupy 1 in 2 solution, but with 0.5 g. in 10 ml. of acetone, sample A showed a considerable amount of insoluble matter; B dissolved to a nearly clear solution, but gradually deposited a brown sediment, and C gave a nearly colourless clear solution. The cupric acetate precipitation test was carried out by dissolving 1 g. of the tannic acid in 20 ml. of water and adding 1.5 g. of cupric sulphate dissolved in 40 ml. of water, then 1.7 g. of crystalline sodium acetate in 10 ml. of water, and lastly 0.5 ml. of acetic acid. After 3 or 4 hours the precipitate was collected, washed, dried and weighed. Sample A gave 1.143 g.; B, 1.179 g.; C, 1.163 g. A larger number of samples would have to be examined before deciding on maximum and minimum figures. D. G. H.

Determination of Cobalt in Foods. N. D. Sylvester and L. H. Lampitt. (*J. Soc. Chem. Ind.*, 1940, 59, 57–60.)—Since most existing methods have usually been concerned with materials of low ash-content, so that separation of cobalt from the ash has been unnecessary, the following procedure is recommended for other types of material:—The sample, which should contain 0.001 to 0.01 mg. of cobalt, is ignited at 500° to 550° C. in a platinum or new silica dish, small quantities of sulphuric and nitric acids being used to aid the process. The residue left after evaporation of a solution of the ash in 15 ml. of 50 per cent. hydrochloric acid is dissolved in a mixture of 5 ml. of the hydrochloric acid and 20 ml. of water, the filtered solution and washings are made up to 20 ml. with water, and 10 ml. of a 1 per cent. solution of α -nitroso- β -naphthol in glacial acetic acid are added. The mixture is boiled, left overnight and filtered on a Gooch crucible, the residue is washed with 10 ml. of 5 per cent. acetic acid and transferred (with the filter-pad) to a Pyrex boiling-tube with the aid first of 2 ml. of warm sulphuric acid and then of 2 ml. of warm 60 per cent. perchloric acid. The contents of the tube are heated to destroy organic matter, *i.e.* until a yellow colour (due to chlorine dioxide) is formed and then disappears; 30 ml. of water are then added, and the solution is extracted with 5-ml. portions of a 0.15 per cent. solution of dithizone in chloroform (*cf.* *ANALYST*, 1935, 60, 378) until the absence of colour in the last extract indicates that all the copper has been removed. Ten ml. of a reagent prepared by making a solution of 250 g. of citric acid in water just alkaline with ammonia and diluting to 1 litre, are then added, followed by sufficient ammonia to produce an orange colour with the residual dithizone, and the solution is then re-extracted in succession with 5 ml. portions each of dithizone reagent and chloroform. The combined extracts, which contain the cobalt, are evaporated to remove the chloroform, and the residue is heated on a hot-plate in a Pyrex beaker with 0.5 ml. of sulphuric acid and sufficient perchloric acid (*e.g.* 5 drops) to destroy the organic matter; most of the residual acid is removed by heating until violent fuming ceases, and the colourless residue is used for the colorimetric determination of cobalt by a modification of McNaught's method (*id.*, 1939, 64, 23). Thus, 6 ml. of water, 1 drop of phenolphthalein indicator, sufficient of a 50 per cent. sodium hydroxide solution to make the solution alkaline, and of 50 per cent. hydrochloric acid to make it just acid, are added in this order to the residue, followed by 1 g. of sodium acetate and 2 ml. of a filtered 0.2 per cent. solution of nitroso-R-salt. The solution is boiled gently for 2 minutes, and again, after the slow addition of 1 ml. of nitric acid, for a further 1 minute. It is then cooled, diluted to 10 ml. and filtered through a No. 42 Whatman paper, and its colour evaluated, *e.g.* in the Lovibond tintometer (1-inch cell), or by means of its extinction-coefficient as determined in the Pulfrich photometer against water (2-cm. cell, S. 53 filter). The results may be read from a graph compiled from data obtained with a standard solution of cobalt (0.001 mg. per ml.); thus, 0 (blank), 0.002, 0.006 and 0.010 mg. of cobalt correspond, respectively, with 0.25, 1.30, 3.60 and 5.80 Lovibond red units, and with extinction-coefficients of 0.086, 0.152, 0.326 and 0.505. The latter method is the more accurate, but the sensitiveness of both methods may be increased by the use of a cell which gives a greater depth of colour for the 10 ml. of solution available. If the colour obtained differs only slightly from that due to the blank, the presence of cobalt is not proved conclusively, and it is therefore suggested that the solution should be passed through a column of alumina which adsorbs the red colour but allows the excess of reagent to pass through; this provides a qualitative test for cobalt (sensitiveness, 0.0002 mg.). Merck's alumina was used; it was made slightly greasy by immersion in a solution of a vegetable oil in acetone, and then washed almost free from the oil with acetone. When known quantities of cobalt (0.005 to 0.10 p.p.m.) were added to tea, milk, fruit pulp and coffee, and when 0.01 mg. was added to a solution containing 3.5 mg. of iron and 1 mg. of copper, the recovery was satisfactory. The cobalt-contents of 19 food samples are recorded, *viz.* gooseberry pulp, 0.004; 5 packet teas, 0.12 to 0.19; green leaf tea, 0.13; China tea, 0.15; straight-grade flour, 0.003; wholemeal flour, 0.010; 4 coffee beans, 0.028 to 0.049; 3 shelled cacao beans, 0.33 to 0.41 (Javanese, 0.03); milk, 0.001 p.p.m. The method was also used for the determination of small amounts of cobalt in commercial copper sulphate, after removal of the copper by electrolysis. J. G.

Biochemical

Respiration of Animal Tissue after Freezing in Liquid Air. F. Lynen. (*Z. physiol. Chem.*, 1940, 264, 146-152.)—In an earlier paper it was shown that cooling in liquid air almost completely inhibited the respiration and fermentative power of yeast. Similar results are now reported concerning the effect of liquid air on the respiration of animal tissues, the uptake of oxygen being very considerably reduced after freezing. The reason for this appears to be that at such low temperatures, the cell-structure is destroyed and some of the soluble constituents of the respiratory system suffer excessive dilution. On the other hand, certain enzyme systems that consist entirely of insoluble components are not affected when the tissues are frozen in liquid air. An example of such a system is that responsible for the dehydrogenation of succinic acid to fumaric acid, with atmospheric oxygen as hydrogen acceptor. The components of this system are succinate dehydrogenase, cytochromes *a*, *b* and *c*, cytochrome oxidase and another factor not yet characterised, and none of these is soluble in water. When sodium succinate was added to the tissue suspension, a marked increase in the oxygen uptake was observed. With heart, liver and kidney, the oxygen uptake was the same whether the tissue was fresh or frozen, whereas with testicle, lung, spleen and Jensen sarcoma, the frozen tissue gave a considerably smaller value than the fresh tissue. This is attributed to a deficiency of these organs in one or other of the components of the enzyme system, but the nature of the limiting factor in each instance is not yet known.

F. A. R.

Determination of Reducing Groups with Porphyrindin, with Special Reference to Egg Albumin. E. Brand and B. Kassell. (*J. Biol. Chem.*, 1940, 133, 437-444.)—The blue dye, porphyrindin, was introduced by Kuhn and Desnuelle (*cf. ANALYST*, 1938, 63, 200), for the determination of sulphhydryl groups in proteins. As solutions of the dye are relatively unstable above 0° C., all estimations were carried out at 0° C. Porphyrindin (5 to 7 mg. per ml.) was dissolved in 0.2 *M* phosphate buffer solution, pH 7.2, at 0° C., and the solution was filtered. It was standardised by titrating 2 ml., in a test-tube kept at 0° C., with an aqueous solution of metal-free cysteine hydrochloride (about 50 mg. per 100 ml.) to the disappearance of the blue colour. The concentration of the cysteine hydrochloride solution is established photometrically or iodometrically; 1 mg. of cysteine is equivalent to 1.16 mg. of porphyrindin. Alternatively, porphyrindin solutions can be standardised iodimetrically, 2 ml. being added to 3 ml. of a solution of 0.3 g. of potassium iodide in 0.5 *N* hydrochloric acid, and the liberated iodine titrated with 0.01 *N* sodium thiosulphate solution. One ml. of 0.01 *N* sodium thiosulphate solution is equivalent to 1.40 mg. of porphyrindin. Cysteine and glutathione are quantitatively oxidised to the corresponding -S-S- compound by the dye solution, and guanidine hydrochloride has no effect on the reaction. The *l*-forms of cystine, cysteic acid, tryptophane, hydroxyproline and histidine, and the *dl*-forms of methionine, serine, phenylalanine and threonine do not reduce porphyrindin. Tyrosine is oxidised by porphyrindin at 0° C. and pH 7.2, with the formation of a pink oxidation product, the reaction being unaffected by guanidine hydrochloride. Native egg albumin does not decolorise the dye, but in heat denatured egg albumin, -SH groups (but not tyrosine) are oxidised. In egg albumin dispersed by guanidine hydrochloride, -SH groups and phenolic groups are oxidised.

F. A. R.

Determination of Neutral Fat Glycerol in Blood with Periodate. L. Voris, G. Ellis and L. A. Maynard. (*J. Biol. Chem.*, 1940, 133, 491-498.)—The principle of the method consists in the isolation of the neutral fat glycerol from the saponified acetone-soluble blood lipids and oxidation of the acidified aqueous solution of glycerol with potassium periodate solution (to give 2 mols. of formaldehyde and 1 mol. of formic acid). *Determination of glycerol with periodate.*—The reagent consists of a solution of 0.625 g. of pure potassium periodate in 500 ml. of 0.1 *N* sulphuric acid. Ten ml. of this solution are added to the aqueous glycerol solution (containing not more than 2.5 mg.), and the mixture is allowed to stand for 20 to 30 minutes. The excess of reagent is determined by titrating the iodine liberated on addition of potassium iodide at pH 4.4 to 7. The mixture is first neutralised, after addition of 3 drops of 15 per cent. magnesium sulphate solution, by adding dilute sodium hydroxide solution dropwise until a faint cloudiness appears, followed by 0.1 *N* sulphuric acid until the turbidity disappears. The reaction mixture is then treated with 10 ml. of a phosphate buffer solution (12 g. of disodium phosphate (hydrate) and 4.2 ml. of 10 *N* sulphuric acid in 100 ml.). Two ml. of 5 per cent. potassium iodide solution are added, and the liberated iodine is titrated with 0.00435 *N* sodium thiosulphate solution, with starch solution as indicator. One ml. of 0.00435 *N* sodium thiosulphate solution is equivalent to 0.5 mg. of potassium periodate or 0.1 mg. of glycerol. *Determination of neutral fat glycerol in bovine blood.*—The phospholipids are precipitated from 100 to 200 ml. of the alcohol-ether extract of plasma by the method of Ellis and Maynard (*J. Biol. Chem.*, 1937, 118, 702), and the acetone solution is evaporated. The residue is saponified, and the solution is acidified with sulphuric acid to liberate the fatty acids, which are removed by extraction with petroleum spirit. The aqueous solution is used for the estimation of glycerol. Alternatively, 5 to 15 ml. of blood plasma are pipetted into 30 ml. of acetone, and 10 g. of anhydrous sodium sulphate are added. The acetone solution is removed, and the residue is re-extracted several times. The combined acetone extracts are evaporated, and the residue is saponified as described above. Good recoveries from added triolein were obtained.

F. A. R.

Stabilisation and Determination of Pyruvic Acid in Blood. E. Bueding and H. Worts. (*J. Biol. Chem.*, 1940, 133, 585-591.)—A significant decrease in the pyruvic acid content of blood occurs on standing, even for a minute, at room temperature. Sodium monoiodoacetate in a concentration of 0.2 per cent. prevents this loss, but produces an increase of 3 to 20 per cent. if the mixture is allowed to stand for 30 minutes; no significant change, however, occurs, in 3 minutes. A 50 per cent. solution of iodoacetic acid in water is adjusted with sodium hydroxide solution to pH 7.8, and a measured volume, corresponding to 25 mg. of iodoacetic acid, is transferred to a bottle containing 20 mg. of dried potassium oxalate. About 5 ml. of blood obtained by venipuncture are caught directly in this bottle. Three ml. of this mixture are transferred dropwise into a flask containing 12 ml. of 10 per cent. trichloroacetic acid solution constantly shaken. After standing for 30 minutes the solution is filtered, and 3 ml. of the filtrate are transferred to a test-tube and treated with 1 ml. of 0.1 per cent. 2,4-dinitrophenylhydrazine in 2 *N* hydrochloric acid. The mixture is allowed to stand at room temperature for at least 10 minutes and then extracted with 4 ml. of ethyl acetate, the layers being mixed with a capillary pipette through which a slow current of nitrogen is blown. The solution is re-extracted twice with 2-ml. portions of ethyl acetate, and the combined extracts are extracted with three 2-ml. portions of sodium carbonate solution. The combined sodium carbonate extracts are extracted with 1 ml. of ethyl acetate, transferred to the cell of an Evelyn colorimeter, and mixed with 4 ml. of 2 *N* sodium hydroxide solution. After 10 minutes the colour, due to pyruvic acid 2,4-dinitrophenylhydrazone, is measured with filter 520. The amount of pyruvic acid is calculated by reference to a standard curve prepared with pure pyruvic acid. F. A. R.

Chlorophyllase. C. A. Weast and G. Mackinney. (*J. Biol. Chem.*, 1940, 133, 551-558.)—Chlorophyllase is an enzyme concerned with the hydrolysis of the phytol ester linkage in chlorophyll, and is capable of operating in high concentrations of alcohol and acetone. An alcohol concentration of 80 per cent. was found to be optimal, whilst the concentration of acetone may be varied from 40 to 70 per cent. The optimal temperature in both instances is 25° C. Water is most effective at 75° C. The activity of the enzyme varies with its source. No activity was found with wild oats (*Avena fatua*); spinach showed high activity in water, but little in acetone or alcohol; figwort (*Scrophularia californica*) showed extremely high activity in alcohol, but none in water; whilst other plants investigated showed intermediate activities. Activity was detected by the formation of characteristic crystals of ethyl chlorophyllide when a leaf section was exposed to the vapour of ethyl alcohol, but the method was found to be of limited value. Two quantitative methods were therefore devised. In the first, 80 per cent. acetone extracts were prepared, and the pigments were partitioned between 80 per cent. acetone and petroleum spirit. The percentage hydrolysed was calculated by spectrophotometric measurement of the original solution and of the acetone solution after partition. In the second method, the pigments were partitioned between 80 per cent. acetone and petroleum spirit, and the pigments in the acetone extract were transferred to ether. Both fractions were evaporated, the residues were saponified with methyl alcoholic potassium hydroxide solution, and ethereal solutions of the unsaponified matter were extracted with 5 per cent. hydrochloric acid. The chlorin *a* and rhodin *g* thus formed were estimated spectrophotometrically. The evidence indicates that the enzyme acts on extracted chlorophyll in 80 per cent. alcohol, but on a denatured chloroplastin in hot water. F. A. R.

Experimental Avitaminosis A in Man. K. H. Wagner. (*Z. physiol. Chem.*, 1940, 264, 153-188.)—Ten volunteers lived for a period of 6 months on a diet almost completely free from vitamin A and carotene, though otherwise adequate. A steady increase in weight was observed in every individual during the first four months of the test, but afterwards all (with one exception) showed a striking decrease in weight. Similarly, at the end of 6 months, marked symptoms of night-blindness were revealed in tests with a Nagel adaptometer; whereas normal persons give readings of about 130,000, the individuals receiving the diet free from vitamin A gave readings between 3500 and 6000, indicating that they were 20 to 30 times less sensitive than normal individuals to the same amount of light. Attention was directed by Jess to a difference between the field of vision of normal persons and of those suffering from night-blindness. Normally, the field of vision for the different colours decreases in the order—blue, yellow, red, green, but in those suffering from night-blindness the order is—blue, red, yellow, green. This same order was also found at the end of 6 months for all 10 persons undergoing test, except that the yellow field tended to fall within the green in one or other half of the eye, and in certain instances even lay wholly within it. This is regarded as a very characteristic sign of vitamin A deficiency in humans, since on administration of vitamin A, the yellow field gradually expanded to its original position between the blue and the red. An examination of the blood of the persons under test was also carried out. Decreased values were found for haemoglobin content, erythrocytes, colour index, thrombocytes and leucocytes, whilst the clotting time was abnormally high. At the end of 6 months, 5 of the individuals were given a supplement of β -carotene, and the other 5 a supplement of vitamin A (vogan). In both instances, the initial doses were too small to effect an improvement in the clinical symptoms (adaptometer reading and blood picture), and the doses were therefore progressively increased until improvement took place. The minimum daily dose of vitamin A required to effect this improvement was 2000 I.U., the average dose being 2500 I.U. Similarly, the average daily dose of β -carotene was found to be 5000 I.U. With these supplements the condition of all 10 persons gradually improved, the weight increased,

and adaptation, field of vision and blood picture all became normal. Estimations were then made of the amounts of vitamin A and carotene in the blood (the former by the Carr-Price reaction on the unsaponifiable matter of the blood serum, and the latter colorimetrically after chromatographing the unsaponifiable matter). In this way, a correlation was found between the intake on the one hand of vitamin A and its concentration in the blood, and on the other hand of β -carotene and the concentration of β -carotene and vitamin A in the blood; the vitamin A was found to make its appearance some 3 weeks after the β -carotene. From estimations of the vitamin A and carotene contents of faeces, it was concluded that about 50 per cent. of the carotene ingested is excreted unchanged, thereby confirming the finding that twice as much β -carotene as vitamin A is required for humans. The results establish the fact that, apart from the effect of absorption, β -carotene and vitamin A are utilised equally efficiently. F. A. R.

Estimation of Tocopherol in Animal Organs. P. Karrer, W. Jaeger and H. Keller. (*Helv. Chim. Acta*, 1940, 23, 464-465.)—The finely minced tissue was twice extracted with 4 volumes of cold alcohol, the first extraction being for 3 hours and the second overnight. The dehydrated tissue was pressed to remove solvent and was then extracted twice with 4 volumes of a mixture (1:1) of alcohol and benzene, using the same extraction times as before. The four extracts were combined, and the solvent was removed by distillation under reduced pressure in an atmosphere of nitrogen. The residue thus obtained was saponified by heating with 8 volumes of 10 per cent. methyl alcoholic potassium hydroxide solution for 1 hour in nitrogen, and the unsaponifiable matter was extracted with peroxide-free ether. The combined extracts were washed with dilute acid and water, dried over sodium sulphate and then distilled under reduced pressure in nitrogen. The vitamin E content of the unsaponifiable matter was estimated potentiometrically by titration with gold chloride solution (*cf. ANALYST*, 1938, 63, 835), and colorimetrically by the method of Emmerie and Engel (*cf. ANALYST*, 1939, 64, 216). The following values were obtained for the α -tocopherol contents in mg. per kg. of tissue:

| | Potentiometrically | Colorimetrically |
|--------------------|--------------------|------------------|
| Horse muscle | 5.308 | — |
| „ heart | 4.892 | 6.17 |
| „ liver | 13.155 | 14.88 |
| „ kidney | 6.25 | — |
| Ox-muscle | 5.852 | 6.21 |
| Ox-liver | 9.540 | 10.55 |
| Pig-fat | 2.185 | 1.97 |

F. A. R.

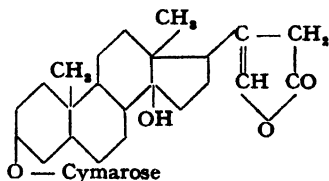
Toxicological

Action of Mustard Gas on Foods. A. Hasskó. (*Tierärztl. Rdsch.*, 1939, pp. 131-133; *Z. Unters. Lebensm.*, 1940, 79, 206.)—Foodstuffs impregnated with mustard gas lose their poisonous qualities the more rapidly the higher the temperature. Bread, bacon, and other dry foods treated with 0.1 ml. of mustard gas are poisonous for at least 20 days at 12° C., and for only 6 to 8 days at 20° C. Flour only moderately impregnated may remain poisonous for more than a month at 16° C. Sausages in "Cellophane" skins withstand the action of the gas for many days, but sausages with skins made from intestines absorb the gas rapidly and lose it only slowly in air. Milk treated with 0.01 ml. of the gas is poisonous after being boiled several times, whereas water treated with 2 ml. of the gas per 10 ml. of water is drinkable after several filtrations through active carbon. The detection of mustard gas poisoning is difficult with dry foods, but with moist foods brown spots or surface changes are soon visible, cucumber and marrow being particularly sensitive. Green peppers, green beans, peas, pears, gooseberries, apples, lemons, and oranges show visible changes only after 24 hours, beets and radishes only after 3 days. Some types of grapes turn brown. Meat from animals which had been given oral doses of mustard gas was not poisonous. E. M. P.

Toxicological Detection of Cocaine. A. Brüning. (*Z. Unters. Lebensm.*, 1940, 79, 93-99.)—The micro-reactions of cocaine and its decomposition product, ecgonine, were examined to discover the most suitable for extracts obtained by the Stas-Otto procedure. The formation of large crystals is promoted by placing a small grain of sand between the cover-glass and the microscope slide and introducing the reacting liquids from opposite sides of the cover-glass, the mingling of the liquids being observed through a binocular microscope. Control reactions should be made with a 1 per cent. cocaine hydrochloride solution. With aqueous 1 per cent. picric acid solution cocaine forms small crystals in 5 to 10 minutes, and these develop into feathery clusters on standing. With a 3 per cent. gold chloride solution typical fern-like aggregates form very quickly. Unlike the picric acid reaction, this reaction is influenced by the state of purity of the extract, and subsequent ethereal extracts give larger and finer crystals than the first. With Martini's reagent (*Mikrochem.*, 1932, 6, 11; *Abst.*, *ANALYST*, 1933, 58, 57) a 1 per cent. cocaine hydrochloride solution gives a dense precipitate, but with a 0.001 per cent. solution characteristic needle-shaped crystals form in a few minutes. Wagenaar's reagent (5 per cent. of lead iodide dissolved in a 30 per cent. aqueous solution of potassium acetate), which has the advantage over

Martini's reagent that lead iodide does not separate on dilution, is less sensitive but yields similar crystals more readily and from concentrated solutions. After a short time these assemble into tree-like formations. In highly decomposed toxicological material much of the cocaine may have been transformed into ecgonine, which is not extracted by the Stas-Otto procedure. De Jong (*Rec. Trav. Pays-Bas*, 1937, 56, 186, 198) has shown that ecgonine forms with barium chloride a double salt soluble with difficulty in water, but readily soluble in hot alcohol. This property was used to detect ecgonine in somewhat putrid material from a cadaver which had originally contained cocaine. The material was acidified with tartaric acid and extracted with hot alcohol. The usual purification of this extract by evaporation with water and alcohol was repeated several times until, after evaporation of the purifying agents, a colourless solution was obtained. This was acidified, made faintly alkaline with sodium hydroxide solution, repeatedly extracted with ether, and, after evaporation to a quarter of its bulk, again extracted with ether. The aqueous residue was evaporated almost to dryness and treated with 10 per cent. barium chloride solution until no further precipitation occurred. The mixture was evaporated to dryness, and the residue was powdered and exhaustively extracted with hot alcohol. The alcoholic solution was filtered, reduced to a quarter of its original bulk by evaporation, and treated with absolute alcohol containing a little sulphuric acid until no further precipitation occurred. It was then filtered, and the filtrate was neutralised with a drop of dilute ammonia and evaporated to dryness. The ecgonine was extracted from the residue with water. Amelink (*Pharm. Weekbl.*, 1938, 75, 861; *Abst., ANALYST*, 1938, 63, 740) has described the reactions of ecgonine with platonic chloride solution followed by sodium iodide solution, with gold chloride solution followed by sodium bromide solution, with mercuric chloride solution, and with a modified form of Dragendorff's reagent. Picric acid gives no crystals with ecgonine solutions; gold chloride forms rectangular plates. Martini's reagent treated with a crystal of ecgonine gives thick needles which dissolve after a time. With Wagenaar's reagent the crystals form more slowly and are larger. Reinecke's salt was found to be a good reagent for the identification of ecgonine, the best procedure being the introduction of a crystal of the salt into a drop of a 1 per cent. solution of ecgonine or its hydrochloride. Crossed crystals are formed first, and these gradually develop through star-like formations into rhombic plates. During their formation these plates often have serrated edges. To determine the effect of impurity on the reaction, a little impure extract obtained from the Stas-Otto procedure was used. Unlike most of the other reactions, the reaction with Reinecke's salt was quite unaffected by the presence of impurity. With cocaine the reagent gives a precipitate consisting of small spherical particles, but, in spite of its sensitivity, the reaction does not give characteristic crystals. A. O. J.

African Arrow-poisons. I. *Adenium somalense* Balf. fil. M. Hartmann and E. Schlitter. (*Helv. Chim. Acta*, 1940, 23, 548-558.)—A new heart glucoside, to which the name somalin has been given, was isolated from the root of *Adenium somalense* (Fam. *Apocynaceae*). In spite of its close chemical affinity to digitoxin, pharmacologically it more nearly resembles strophanthin. It comprises a molecule of digitoxigenin and a molecule of cymarose, and has the formula:



F. A. R.

Bacteriological

Action of Sulphanilamide. (*Brit. Med. J.*, 1940, I, 775-6.)—There have been several conceptions of the possible mode of action of sulphanilamide on *Streptococcus pyogenes*, including that of the neutralisation of toxins, the stimulation of leucocytic activity, and antibody formation. What appears to be a complete solution of the problem has now emerged from the work of Paul Fildes and his collaborators. This was preceded by a search by D. D. Woods for a substance that would antagonise the bacteriostatic action of sulphanilamide *in vitro*—i.e. would furnish the bacterial cell with something, essential for its growth, of which sulphanilamide deprived it; he found that such a substance was contained in yeast. T. C. Stamp and H. N. Green found that it was also contained in an extract of *S. pyogenes* and *Brucella abortus*. By using completely synthetic media, every constituent of which was known, the chemical constitution of this substance was deduced step by step, and the conclusion reached that it is *p*-aminobenzoic acid. Confirmation of this is afforded by the fact that pure *p*-aminobenzoic acid neutralises the antibacterial action of sulphanilamide and sulphapyridine, so that mice inoculated with *S. pyogenes* and given *p*-aminobenzoic acid are not protected by sulphanilamide. *S. pyogenes* is capable of elaborating this substance from media which do not contain it, provided that the inoculum is sufficiently large; it is presumed that the effect of sulphanilamide is to inhibit the

enzyme action by which further utilisation is made of this substance and thus arrest growth. Fildes has shown that an exact parallel is to be found in the bacteriostatic action of low concentrations of mercuric chloride. In all but low concentrations mercuric chloride is of course bactericidal, but in low concentrations inhibition of growth is due to the removal of -SH compounds that are essential for bacterial metabolism. This is proved by the fact that growth can be restored by adding glutathione, a quantitative relation existing between the amount required and the amount of mercuric chloride present.

D. R. W.

Water

Application of Hydrogen Ion Concentration to Boiler Water Treatment. G. W. Bond. (*J. Chem. Met. and Mining Soc., S. Africa, 1939, 40, 59-68.*)—The following pH values for different solutions of commercial chemicals used for the treatment of water throw light on their behaviour in the process:

| | pH | | |
|---------------------------------------|--------------|---------------|----------------|
| | 10 per cent. | 0.4 per cent. | 0.04 per cent. |
| Ferrous sulphate (copperas) | 3.0 | 3.8 | — |
| Alumina, ferric (local) | 2.0 | 3.2 | — |
| Filter alum (imported) | 2.8-3.0 | — | — |
| Soda ash (sodium carbonate) | 11.6 | 11.6 | 11.6 |
| Sodium aluminate (Alfloc brand) .. | 11.6 | — | — |
| Caustic soda | 12.0 | 12+ | 12+ |
| Trisodium phosphate (crystalline) .. | 12.0 | 12.0 | 12.0 |
| Disodium phosphate (crystalline) .. | 9.5 | 9.4 | — |
| Monosodium phosphate (crystalline) .. | 4.4 | 4.53 | — |
| Sodium hexa-metaphosphate ("Calgon") | 5.8 | 6.25 | 7.0 |
| Lime water (saturated solution) .. | | over 12 | |

Generally speaking, and within strict limits, the higher the pH the greater the amount of oxygen that can be tolerated in feed-water. The solvent action of acids on iron may be represented as: $\text{Fe} + 2\text{H}^+ \rightleftharpoons \text{Fe}^{2+} + \text{H}_2$. The direction taken by this reaction will be determined by the pH of the water and the electrode potential of iron, together with its hydrogen overvoltage. The more easily the hydrogen film is removed from the surface the more readily will the reaction proceed to the right, i.e. the more easily will the metal be corroded, and any hydrated oxide film deposited on the surface will have a protective action and thus inhibit corrosion. In absence of oxygen corrosion of iron is proportional to the hydrogen ion concentration to pH 8.4, beyond which evolution of hydrogen almost ceases. In actual practice it is difficult to work up to this figure without getting excessive alkalinity in the boiler water. The acids that act in boiler feed waters are carbonic acid and, possibly, hydrochloric acid derived from the hydrolysis of magnesium chloride. Decomposition of sodium carbonate in high-pressure boilers is a source of carbon dioxide and may lead to corrosion of turbine blades and to formation of an acid condensate. To counteract this, alkali must be added in the form of lime, caustic soda or trisodium phosphate; the use of lime, however, may lead to scale problems. It is also necessary to add alkali to counteract the potential corrosiveness of sea water, or waters rich in magnesium chloride or nitrate. In coastal stations there may be small leakage of sea water used in the cooling system into the boilers. With regard to the controversial problem of caustic embrittlement, many chemists doubt very much if this is really due to excessive causticity, and the recent work of Straub himself (*Mechan.*

Engineering, 1938, 60, 371), the originator of the now famous A.S.M.E. ratio of $\frac{\text{Na}_2\text{SO}_4}{\text{Na}_2\text{CO}_3}$, throws doubt on the efficacy of that ratio. In any event, a large excess of alkali is undesirable for other reasons. When testing the pH of feed water it is essential to fill the sample bottle completely with the water from the sampling cock, and to make the determination as soon as the stopper is removed. Even a comparatively short exposure to air may cause a discrepancy in pH of as much as 0.5.

Practical Methods for Sterilisation of Water consumed by Evacuated Populations. R. D. de la Rivière. (*Mouvement Sanitaire*, 1939, 16, 455-465; *Bull. Hyg.*, 1940, 15, 117-8.)—Evacuated populations (of France), communities which shelter them, and troops in the neighbourhood are in danger of contracting infectious, and especially water-borne diseases. Where there is a reliable system of distribution (and all "concessionaires" are responsible by law for the purity of the water they supply) increased vigilance only is necessary. Where new distributing installations are adopted it will probably be simpler and cheaper to depend on chlorination rather than ozonisation for sterilisation. Two methods are successfully employed in various French towns:—"Javelisation" and "Verdunisation." The Conseil Supérieur d'Hygiène points out that for the former the water must be limpid and contain not more than 3 mg. of organic matter per litre with no appreciable amounts of ammonia, urea, nitrites or iron salts. The importance of adequate control of the amount of chlorine and time of treatment is emphasised. Where there

is no supply of pure water by main three means of dealing with the problem are available:—

(1) Water sterilised elsewhere may be imported by tank-wagons and stored, as in 1914, in reservoirs of reinforced concrete. According to Manceau the water should not be used until 2½ hours after chlorination or stored for more than 12 hours. (2) Water available locally may be drawn and treated, as, for instance, by motor pumps fitted with an automatic Verdunising system, such as were successfully employed by Bunau-Varilla on the Verdun front in 1915. There are also at present on trial sterilisation plants mounted on trailers with capacity of 500 litres and possible daily output of 8000 litres. In these the "carbo-chlore" method of Gambier is adopted, in which the water is treated for 10 to 30 minutes with 10 mg. of chlorine per litre and filtered through activated charcoal. In addition, there are pinnacles with sterilising plants and storage for treating river water. (3) Disinfection of drinking water may be carried out by the consumers; boiling is simple and effective, or, alternatively, one drop of *eau-de-Javel* may be added to a litre of water and, ten minutes later, a tablespoonful of wine or cider to neutralise excess of chlorine and mask any taste. In general, the use, by the layman, of filters and chemical agents on the market, is inadvisable, but Tanon's method may be used. For this, two solutions are employed:—(A) Iodine, 1 g.; potassium iodide, 2 g.; water, 200 ml.; and (B) Sodium thio-sulphate, 10 g.; water, 50 g. To a litre of the water to be tested solution A is added, drop by drop, until a faint brownish tint is obtained; if this fades before 20 minutes, 2 more drops are added. After 20 minutes one drop of solution B is added.

D. R. W.

The Presence of Sulphide in (Harrogate) Mineral Water and its Oxidation by Air. A. Woodmansey. (*Harrogate Spa Medical J.*, 1940, 2, [ii], 8.)—The freshly-drawn water contained 9 parts per 100,000 of sulphide sulphur. When kept in a partly-filled stoppered bottle for 1 to 2 weeks, the content of sulphide sulphur gradually dropped to nil, the sulphide becoming oxidised by air to sulphur, thiosulphate and sulphate, and a little being lost as hydrogen sulphide. The loss of hydrogen sulphide is more pronounced when the water is poured from one vessel to another. The oxidation of the sulphide is promoted by the water being slightly alkaline; the oxidation was much less rapid when the water was slightly acidified. The probable source of the sulphide in Harrogate water is a deep-seated mass of igneous rock, and no evidence exists of any connection with the surface of a volcanic vent.

S. G. C.

Agricultural

Selenium in Canadian Wheat. J. G. Malloch. (*Food Manufacture*, 1940, 15, 160.)—The Director of the National Research Council of Canada points out that previous work of Byers and Laken may have caused apprehension that selenium may be present in dangerous amounts in some parcels of wheat exported from Canada. An extensive investigation by Thorvaldson and Johnson (to be published in the *Canad. J. Research*) has shown that the plants analysed by Byers and Laken were mostly of the so-called "indicator" species, which requires selenium for proper development. Only 4 samples of seedling wheat plants and none of grain were included. In Thorvaldson's investigation 2230 samples of wheat grain were examined, sufficient to show the conditions in the main wheat-producing area of Western Canada. The growth of rats is adversely affected by 6 p.p.m. of selenium, but not by diets containing less than 3 p.p.m. In the wheat samples examined the highest concentration was 4 p.p.m., but this was the only sample containing more than 3 p.p.m. The average amount was less than 0.5 p.p.m. It is highly probable that the selenium content of the wheat exported approximates to this figure because of the great amount of mixing with wheat from different areas that occurs before the grain reaches the seaboard.

Organic

Identification and Determination of Phenolic Compounds. L. F. Levy. (*J. South African Chem. Inst.*, July, 1939, 1-8.)—The following reagents are used:—(i) A 0.03 per cent. solution of 2 : 6-dibromo-4-aminophenol chlorostannate in 95 per cent. alcohol (*cf. "Organic Synthesis,"* 15, 8). This is permanently stable in a brown bottle. It contains a small amount of hydrochloric acid, for which the stock buffer must be compensated by addition of alkali. (ii) Neutral sodium hypochlorite solution (Dakin).—A mixture of 100 ml. of 10 per cent. (w/v) calcium hypochlorite solution and 100 ml. of a solution of sodium carbonate (4.5 g.) and sodium bicarbonate (4.8 g.) is filtered after a few hours. Immediately before use, the solution is diluted to 0.05 per cent. of sodium hypochlorite (strength found by titration with *N*/10 sodium thio-sulphate solution). (iii) *Stock solutions.*—(0.1 per cent.) of phenol, *o*-cresol and *m*-cresol. These are diluted to 0.001 per cent. immediately before use. *Buffer solutions.*—(iv) (Clark and Lubs) pH 9.8—Fifty ml. of a solution of boric acid (12.4 g. per litre) and potassium chloride (14.9 g. per litre) are mixed with 16.3 ml. of *N*/2 sodium hydroxide solution and diluted to 200 ml. v) (Sørensen) pH 10.5—Boric acid (12.4 g.) is dissolved in 196 ml. of *N* sodium hydroxide

solution and diluted to 1 litre. (Tested with aniline yellow GG, at pK assumed to be 11.0.) **Identification.**—To a buffered solution of phenols (pH 9.2 to 10.5) mixed with a little (i) is added (ii) in the ratio of 2 mols. to 1 of (i). The indophenol blue colour develops in $\frac{1}{2}$ to 3 minutes (*o*-cresol), 1 to 3 minutes (*m*-cresol), or 5 minutes (phenol). Acidification with acetic acid to pH 3-4 produces a colour characteristic of the phenol present, e.g. phenol, deep reddish-violet; *o*-cresol, light brown; *m*-cresol, reddish-violet; *p*-cresol, no reaction; xylenols, deep bluish-violet. In a distillate containing cresols colour development after 3 minutes indicates the presence of phenol. **Determination.**—Interfering substances in the test solution must be removed by steam distillation. The distillate is buffered to approximately pH 9.8 (indicator, thymol-phthalein), and the phenols are determined colorimetrically in Nessler tubes (100 ml.). Five ml. of (iv), the test solution, containing approximately 0.1 mg. of cresol, and 1 ml. of (i) are diluted to a definite volume; a comparison solution similarly prepared, and 1 ml. of (ii) are mixed with each, and the colours are compared after 3 minutes.

For mixtures of *o*- and *m*-cresols a comparison solution similarly mixed is required. The method must be modified for phenol, which is oxidised rapidly at pH 10.5. Beakers (250 ml.) replace the Nessler tubes, the mixtures are made up to 110 ml., and the hypochlorite solution is added (from watch-glasses) simultaneously to each beaker. After mixing, 100-ml. Nessler tubes are filled to the mark with the solutions, and the colours are compared in the fifth minute (it is essential that comparison shall be made within 6 minutes, and the pH of the solutions must be as close as possible). When adjustment is difficult, a modification of the Houghton and Pelly method enables a large number of determinations of phenol to be made with the same reagent, as follows:—0.1 g. of *p*-nitrosodimethylaniline is dissolved in 2 ml. of 95 per cent. alcohol, diluted to 100 ml. with water and reduced by excess of zinc dust followed by constant shaking with hydrochloric acid, added drop by drop, until the colour disappears. After precipitation of the dissolved zinc by sodium bicarbonate, the reagent is stable for hours if kept in an atmosphere of carbon dioxide. (For determination, cf. ANALYST, 1937, 62, 117). In a mixture of cresols only, the *o/m* ratio may be found as in pH determinations with two colour indicators, the *o*- and *m*-cresols as described above, and the total (*o*-, *m*-, and *p*-) cresols by Chapin's method (cf. J. Biol. Chem., 1927, 72, 649); hence each cresol may be determined.

Detection of phenols.—To 10 ml. of a solution, slightly alkaline to litmus, are added successively 5 ml. of 2 per cent. borax solution, 3 drops of (i) and a small drop of stock (undiluted) hypochlorite solution; the blue colour forms immediately. On addition of 2 drops of acetic acid the characteristic phenol or cresol colour appears. E. B. D.

Identification of Groups of Dyestuffs by their Reaction with Ferrous Hydroxide. H. Eichler. (Z. anal. Chem., 1940, 119, 91-94.)—Solutions of azo or triphenylmethane dyes are decolorised on addition of ferrous hydroxide, which is converted into ferric hydroxide. The azo dyes are decomposed into their component primary amines, the course of the reaction being

$$4Fe(OH)_2 + R.N:N.R' + 4H_2O \rightarrow R.NH_2 + R'.NH_2 + 4Fe(OH)_3$$

With triphenylmethane dyes the reaction is more complex, and has not yet been elucidated. The reaction is carried out as follows:—Insoluble matter is removed from the dye solution by filtration and leuco bodies are oxidised by means of a current of air. The solution is then treated with a 30 per cent. solution of sodium, potassium or ammonium hydroxide and with saturated ferrous sulphate solution. The mixture may be boiled for a short time to promote settling of the precipitate, after which the clear liquid is decanted. Mono- and poly-azo dyes containing sulpho and carboxyl groups are completely destroyed, precipitate and solution being free from dye. In this group the following dyes were tested:—Methyl orange, methyl red, cochineal scarlet PS, cloth red 3B, crocein scarlet 7B, fast red A, toluylene orange R and N, Congo red, azo blue, orange R, palatine red A, rosazurine G, benzo brown, cloth red Bo, benzo fast scarlet SBS, azo-grenadine S, oxamine blue B, trona red 3B, mikado orange R, "Plutoschwarz A," toluylene brown BBo, "Plutoschwarz R and F" cloth red 3G, and azofuchsin GN. With the following azo dyes containing neither sulpho nor carboxyl groups the colour was either not destroyed or incompletely destroyed and was present in the solution and in the precipitate:—Chrysoidine G and T, Vesuvine OOO extra, Bismarck brown GOOO, and aniline yellow. With the following triphenylmethane dyes the solution and precipitate were free from colour:—New patent blue B conc., brilliant green O, methyl green, fuchsin, methyl violet 3RO and 5B conc., new Victoria blue B, new patent blue R, brilliant green extra, wool green BS, new green RS, eosin GO, new green extra, acid fuchsin NS, night blue, light green, rosolic acid, phenolphthalein, turquoise blue G, cyanol, Victoria blue R. With the following triphenylmethane dyes containing sulpho groups the solution was colourless, but the colour reappeared in the solution of the precipitate in dilute mineral or acetic acid:—acid fuchsin, alkali blue 7B, water blue, rhodamine 6G, new fuchsin, Poirrier's blue C4B. With the triphenylmethane dye auramine O, the precipitate contained dye and the colour reappeared in the filtrate when it was acidified and exposed to air. Iron lakes were formed with gallein and the oxyanthraquinone dyes. Acid anthraquinone dyes (alizarin, cyanine green K, anthracyanine FLA, alizarin saphirole A) formed lakes, and the colour reappeared in the alkaline solution when it was exposed to air. With the phenosafranin dyes (safranin G, indoine blue R and azo carmine G) the colour reappeared in the alkaline or acidified solution when it was exposed to the air, and the precipitate contained dye. With rosinduline dyes (phenyl rosinduline and Magdala red) the solution had no colour even on oxidation, but

the dye reappeared when the precipitate was dissolved in dilute acid. With thiazine dyes (methylene blue and methylene green) the colour reappeared in both acid and alkaline solutions and was present in the precipitate. With the oxazines (new blue R and celestine blue R) the colour returned to the solution and was present in the precipitate. With the oxazones (resorufin, resazurin, iris blue and tetrachlororesorufin) the colour returned to the alkaline solution exposed to air and was present in the precipitate. With the indophenols (resorcin indophenol) the colour reappeared in the alkaline solution exposed to air. Acridine dyes (benzoflavin, phosphine and rheonine A) gave a colourless solution and dye in the precipitate. Quinoline yellow occurred unchanged in the precipitate and solution. Nigrosine B and other nigrosines gave a colourless solution and dye in the precipitate. The pyrazolone dye tartrazine gave a sparingly soluble lake in acid solution. Nitro dyes (picric acid) gave metallic lakes. Oxyketone dyes (galloflavine) gave iron lakes decomposed by acids with liberation of the original dye. With fluorindines the colour reappeared on exposure of the alkaline solution to the air. A. O. J.

Estimation of Viscose Rayon. R. W. McKay. (*Amer. Dyestuff Rep.*, 1940, 29, 25-28; *J. Soc. Dyers and Col.*, 1940, 56, 243.)—For the estimation of viscose rayon in admixture with cotton, wool or silk, the fabric is cut into small pieces not exceeding 0.2 cm. in diameter and treated for an hour at -3°C . with 2*N* sodium hydroxide solution. The viscose rayon dissolves whilst cotton, wool and silk are hardly affected. The residue is washed with chilled 2*N* sodium hydroxide solution, then with water and finally with a little 0.2*N* hydrochloric acid, after which it is dried and weighed. The results are accurate to the nearest unit per cent. Resin finishes may interfere with the analysis.

Inorganic

Colorimetric Determination of Small Amounts of Lead by means of Dithizone. H. Fischer and G. Leopoldl. (*Z. anal. Chem.*, 1940, 119, 161-188.)—Recent work on the subject is reviewed and various investigations by the authors are discussed. **Extraction of Lead.**—The preferred method for amounts of lead of the order of 1 to 25 γ involves treating the solution with dilute ammonia until it is just alkaline to litmus paper. When elements precipitated by ammonia are present, sufficient alkali citrate or tartrate should previously be added to prevent this. Potassium cyanide is added to convert interfering metals into complex cyanides; usually 3 to 5 ml. of 5 per cent. potassium cyanide solution to 10 ml. of the test-solution are sufficient, but, where justified by the presence of appreciable amounts of, for example, zinc, copper, silver or mercury, considerably increased amounts of potassium cyanide may be introduced without detriment. To overcome the interfering effect of oxidising substances which may be present, such as ferricyanide, a little hydroxylamine hydrochloride or sodium hydrosulphite is added, and the solution is boiled for a short time and then cooled to room temperature. It was found that small amounts of copper catalyse the oxidising action of various substances on lead dithizonate, but the treatment just mentioned eliminates this. Special adjustment of the pH of the solution between 8 and 10 is unnecessary, as the degree of alkalinity provided by the excess potassium cyanide allows of complete extraction of the lead. The solution, contained in a separating funnel, is well shaken with 3 to 5 ml. of green dithizone solution (about 6 mg. of dithizone in 100 ml. of carbon tetrachloride); the carbon tetrachloride layer containing the red lead dithizonate is drawn off, and the process is repeated until the non-aqueous portion is no longer red. **Colorimetric Determination of Lead.**—Advantages are offered by the following "mixed-colour" process. The combined extracts, obtained as described above, are shaken with 5 ml. of 1 per cent. hydrochloric acid. The green non-aqueous layer containing the dithizone is separated and rejected. The aqueous portion, containing the lead as chloride, is washed free from dithizone by shaking with a little carbon tetrachloride, and then transferred to a 25-ml. stoppered measuring cylinder and diluted to 10 ml. with 1 per cent. hydrochloric acid; 2 ml. of ammoniacal cyanide solution (75 ml. of ammonia, sp. gr. 0.9, with 100 ml. of 10 per cent. potassium cyanide, diluted to 1 litre) are added. Dithizone solution is added little by little from a burette, with occasional shaking, until there is an excess over that required to give the full red colour with the lead present, a "mixed-colour" between red and green being produced. (If the amount of lead is unexpectedly high, and a "mixed colour" cannot therefore be obtained, a fresh start should be made with a smaller sample). Into another similar stoppered cylinder are introduced 10 ml. of 1 per cent. hydrochloric acid, 2 ml. of the ammoniacal cyanide solution and the same volume of dithizone solution as was added to the sample solution. The liquid is titrated with standard lead solution, with shaking, until the colour matches that of the sample; the amounts of lead in the two liquids are then the same. The process is suited to amounts of lead from 1 to 25 γ . **Determination of Lead in presence of Bismuth.**—The test-solution, containing not more than 5 mg. of bismuth, is treated with citrate and made slightly ammoniacal. An equal volume of 10 per cent. potassium cyanide solution and 2 ml. of dithizone solution are added, the liquid is well shaken mechanically for 5 minutes and the red carbon tetrachloride layer is separated. The extraction is repeated until the extract shows a pure orange colour. This indicates that lead dithizonate, which forms in preference to bismuth dithizonate (orange colour), has been completely extracted. To verify the completeness of extraction of lead, the last extract is shaken with 1 per cent. potassium cyanide solution, which discharges the colour due to bismuth. The

combined extracts are well shaken with 5 ml. of 1 per cent. hydrochloric acid, the metals passing into the aqueous phase. The small amount of bismuth accompanying the lead is separated by adjusting the pH to 2.8–3.0 (Congo-red indicator paper changing to a bluish tint) and extracting the bismuth by repeated mechanical shaking with small portions of dithizone solution; the solution remains green when extraction is complete. After a final shaking with pure carbon tetrachloride, the aqueous solution is rendered slightly ammoniacal, 5 ml. of 5 per cent. potassium cyanide solution and 1 ml. of 10 per cent. hydroxylamine hydrochloride solution are added, and the lead is extracted and determined as indicated above. Good, but slightly low, results were obtained in tests with 7.4 to 24.6 γ of lead and 5 mg. of bismuth. *Determination of Lead in presence of Thallium.*—Lead is extractable in preference to thallous thallium, but it is not possible to judge the progress of the separation by colour alone, as both lead and thallium dithizonates are red. The expedient of shaking each extract with 0.5 per cent. potassium cyanide solution is suggested; lead dithizonate is unaffected, whilst thallium dithizonate is decomposed. The combined extracts, showing the red colour of the lead compound, are tested colorimetrically for lead-content by the usual "single-colour" process. Slightly high results were obtained in tests with 5 to 19 γ of lead in presence of up to 500 γ of thallium. *Qualitative Tests.*—Stannic tin does not interfere with the detection of lead with dithizone. Stannous tin gives a red colour like that with lead. The interference is partly suppressed by the presence of an excess of 10 per cent. potassium cyanide solution during the extraction process; if a red extract is obtained, it should be shaken with several separate portions of 1 per cent. potassium cyanide solution; any red colour due to stannous dithizonate is discharged. The colour of bismuth dithizonate is similarly discharged by 1 per cent. potassium cyanide solution. It is possible to detect 0.5 γ of lead in presence of 1 mg. of bismuth and 2.5 mg. of stannous tin. An extensive bibliography is appended to the paper.

S. G. C.

Colorimetric Determination of Lead as Chromate with the aid of Diphenylcarbazide. T. V. Letonoff and J. G. Reinhold. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 280–284.)—The process is applicable to a few hundredths of a milligram of lead. The solution, containing hydrochloric acid, is treated with 2 ml. of 20 per cent. sodium citrate solution and sufficient ammonia to give a pink colour with phenol red indicator. Acetic acid is added to produce an orange-yellow colour with the indicator, followed by 1 ml. of 40 per cent. ammonium acetate solution and 1 ml. of 30 per cent. potassium chromate solution. The liquid is kept overnight and centrifuged. The precipitate is washed by centrifuging with several 10-ml. portions of 0.4 per cent. ammonium acetate solution. The precipitate is dissolved in 3 ml. of 10 per cent. hydrochloric acid, 10 ml. of 0.02 per cent. aqueous diphenylcarbazide solution are added, and the red colour is compared colorimetrically with standards prepared similarly by suitable dilution of a solution of lead chromate in 10 per cent. hydrochloric acid (39 mg. of lead chromate in 100 ml.; 1 ml. = 0.25 mg. of lead). The novelty of the method consists in the nature of the precipitate, which, when formed under the conditions stated, producing a pH of 6.6 to 7.4, was proved to be a double chromate of lead. The intensity of colour produced is double that shown by a standard solution of lead chromate containing the same amount of lead. At lower pH values of precipitation, normal lead chromate is precipitated. The method was applied to the determination of lead in biological materials after dry ashing.

S. G. C.

Volumetric Determination of Manganese and Lead Dioxides. C. Mahr and H. Ohle. (*Z. angew. Chem.*, 1939, 52, 618.)—The determination is based upon the oxidation of thiourea by the metallic oxide (*cf.* ANALYST, 1940, 189). *Manganese dioxide* (0.07 to 0.08 g.) is added to a mixture of 5 ml. of syrupy phosphoric acid, 25 ml. of sulphuric acid (1:1), 5 ml. of 1 per cent. potassium iodide solution and 20 to 25 ml. of 0.1 *N* thiourea solution, and the whole is diluted to 75 ml. The solution is gently heated on a water-bath and shaken until the oxide is dissolved, after which it is cooled to 35° C. and the excess of thiourea is titrated with 0.1 *N* bromide-bromate solution and starch indicator until the blue end-point appears. It is then diluted to 250 ml. with water (35° C.) and the titration is resumed until the blue colour returns (ANALYST, 1939, 64, 622).—*Lead peroxide* is determined similarly, the weighed quantity being added to a mixture of 15 to 20 ml. of 60 per cent. perchloric acid, 5 ml. of 1 per cent. iodide solution, and a measured excess of thiourea reagent; the operation is otherwise conducted as described above. W. R. S.

Volumetric Determination of Ferric Iron by Means of Mercurous Nitrate. F. R. Bradbury and E. G. Edwards. (*J. Soc. Chem. Ind.*, 1940, 59, 96–98.)—The advantage of the method is that it utilises a standard solution unaffected by atmospheric oxygen, *viz.* a 0.1 *M* solution of mercurous nitrate in 5 per cent. nitric acid. The titre remains unchanged for two weeks. The neutral or slightly acid solution of ferric sulphate is treated with 40 per cent. ammonium thiocyanate solution, and titrated with the mercury solution until the red colour just disappears. Towards the end the liquid is titrated, drop by drop, during constant agitation, but the whole operation should be conducted with dispatch, as the colour slowly returns on standing. Dilute sulphuric acid has no effect on the titration, but the concentration of hydrochloric acid, if present, should be below 0.1 *N* otherwise a positive error is introduced. The quantity of thiocyanate added should be at least ten equivalents per equivalent of ferric iron. The method is stated to be accurate within 0.1 per cent. Further investigation into the mechanism of the reaction and its practical application is proceeding; so far it is proved that the ferric iron must be present as the thiocyanate complex for reduction to take place.

W. R. S.

Estimation of Cerium in Cerous Salts. J. Plank. (*Magyar Chem. Foly.*, 1939, 45, 100-103; *Chem. Age*, 1940, 42, 294.)—When a concentrated solution of potassium carbonate is added to a solution of a cerous salt it gives a white precipitate of cerous carbonate, which is soluble in excess of the carbonate solution, forming a double salt. The colour of the solution changes to yellow by oxidation of the potassium cerous carbonate to the perceric salt, and the intensity of the resulting colour affords a measure of the amount of cerium present. As other ions interfere with the reaction, the method is applicable only to solutions of pure cerium salts.

Modified Iodate Method for the Determination of Barium. F. C. Guthrie. (*J. Soc. Chem. Ind.*, 1940, 59, 98.)—The solution of barium chloride (0.06 to 0.3 g.) in 20 ml. of water is heated to boiling, stirred, and treated with 25 ml. of a solution of potassium iodate (32 g. per litre) through a dropping tube, which is finally rinsed with small quantities of water. The mixture is boiled for about a minute, allowed to cool, transferred to a 100-ml. graduated flask, made up to volume, mixed, set aside for an hour, and filtered. Aliquot parts of the filtrate are treated with 2 g. of potassium iodide, 5 ml. of 2 N sulphuric acid, and 100 ml. of water, and the liberated iodine is titrated with 0.1 N thiosulphate solution standardised against potassium iodate. Calcium, if present, causes positive errors. The above procedure of partial filtration and indirect titration avoids the washing of the iodate precipitate in the direct titration method (the washed precipitate being treated with sulphuric acid and iodide solution), which gives somewhat low results owing to solubility losses. W. R. S.

Determination of Tellurium in Tin-Rich Alloys by Volatilisation. W. T. Pell-Walpole. (*Publ. No. 96, Int. Tin Research and Dev. Council*, 1940, pp. 3.)—The sample is heated for 1 hour at 1000° C. at 0.02 to 0.03 mm. pressure. Tellurium is lost quantitatively as the compound TeSn , permitting determination by loss in weight. Other volatile elements, such as cadmium, must be absent. S. G. C.

Volumetric Determination of Sulphate. A. Krüger. (*Z. anal. Chem.*, 1940, 119, 216-221.)—The possibility of adapting the reaction $\text{Na}_2\text{SO}_4 + \text{BaCO}_3 = \text{BaSO}_4 + \text{Na}_2\text{CO}_3$ has been investigated. For various reasons it was only found possible to obtain practically theoretical results by adopting a detailed, lengthy and rigid technique. The amounts of sodium sulphate employed were of the order of 1.0 g. S. G. C.

Interference of Sulphite with the Determination of Sulphate by the Tetrahydroxyquinone Method. H. L. Kahler. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 266-267.)—Sulphite interferes, producing high results. Approximately 85 per cent. of the sulphite present is recorded as sulphate. The sulphite should, therefore, be removed by acidifying and boiling the solution. S. G. C.

Determination of Silicon in Fluorspar and Cryolite. T. Nakamura. (*J. Soc. Chem. Ind. Japan*, 1940, 43, 33b.)—The silicon is converted into potassium fluosilicate which is ultimately titrated with alkali. *Fusion process.*—A 1-g. sample is fused with 5 g. of fusion mixture. The cooled melt is dissolved, as far as possible, in water; the liquid is transferred to an Erlenmeyer flask fitted with a 2-holed stopper carrying (1) a tap funnel, the stem of which dips below the liquid, (2) two absorption bulbs of the "potash bulb" type, in series containing respectively 1:1 hydrochloric acid and water. The liquid in the flask is neutralised to methyl orange by admitting hydrochloric acid from the tap funnel. A current of air is passed through the apparatus for 15 minutes to remove carbon dioxide, which passes through the washing bulbs. The contents of the bulbs are then transferred to the solution in the flask, an equal volume of alcohol is added, and the solution is rendered distinctly acid. After 2 hours the precipitate of potassium fluosilicate is filtered off, washed with a 2 per cent. solution of potassium chloride, and titrated with N/10 sodium hydroxide solution, methyl red being used as indicator (Travers, *ANALYST*, 1921, 46, 522). *Hydrofluoric acid process.*—A 1-g. sample, contained in a platinum dish, is allowed to remain in contact with 1 to 2 ml. of hydrofluoric acid reagent for 15 to 30 minutes. (The reagent consists of a mixture of equal volumes of conc. hydrofluoric acid and alcohol, with 2 per cent. of potassium chloride; any precipitate of potassium fluosilicate is filtered off.) The liquid is neutralised with 30 per cent. potassium hydroxide solution, phenolphthalein being used as indicator, and cooled. Equal volumes of alcohol and of 2 per cent. of potassium chloride solution are added, the liquid is acidified, and the precipitated potassium fluosilicate is filtered off and titrated as before. It is noted that cryolite precipitated during the titration adsorbs fluosilicate, rendering it necessary to digest the liquid on a water-bath and to continue the titration until a permanent yellow colour is obtained. S. G. C.

Microchemical

Micro-potentiometric Method of Formol Titration. A. Janke and E. Mikschik. (*Mikrochem.*, 1939, 27, 176-179.)—Formol titration (Sørensen, *Biochem. Z.*, 1908, 7, 45; *Abst.*, *ANALYST*, 1908, 33, 19) is now widely used for the determination of amino nitrogen in amino acids and peptides or the indirect determination of nitrogen in ammonium salts. Potentiometric titration, which has many advantages, especially with coloured liquids, may be applied to as little

as 0.1 ml. of the sample by using a special micro glass electrode as the titration vessel. The electrode is silvered on the outside to make electrical contact with the terminal and packed with cotton-wool in a cardboard holder to prevent damage to the delicate glass membrane. The electrode, which is bulb-shaped, is supported by the neck, and this is protected by a collar of asbestos paper, so that it can be shaken without risk. For the titration 0.1 N sodium hydroxide solution is used with a tapless micro-burette (Schwarz, *Mikrochem.*, 1933, 13, 1; 1935, 18, 106, 309; Abst., ANALYST, 1933, 58, 422) or similar type of burette. The method has been applied to the determination of amino nitrogen in sugar beet. J. W. M.

Micro-gravimetric Separation of Zinc and Uranium. E. Kroupa. (*Mikrochem.*, 1939, 27, 1-7.)—A method of separating zinc from uranium has been worked out in connection with the determination of the lead, thorium and uranium contents of feebly radioactive minerals in the measurement of geological time by the so-called "lead method"; the elements to be determined constitute a few hundredths of 1 per cent. of the minerals. The chloride or nitrate solution containing the zinc and uranium is buffered with monochloroacetic acid and sodium acetate as in Mayr's macro-procedure, the zinc is precipitated as zinc sulphide, the precipitate is dissolved in hydrochloric acid, and the zinc is determined as zinc ammonium phosphate. The filtrate containing the uranium is evaporated and treated with hydrochloric acid and bromine water. The uranium is then determined by precipitation in the usual way with 8-hydroxyquinoline in acetic acid solution containing ammonium acetate. In 11 determinations of uranium in amounts ranging from 0.5 to 12 mg. and of zinc ranging from 0.5 to 8 mg. the differences between the calculated and determined values never exceeded 1 per cent. J. W. M.

Quinaldinic Acid as a Micro-Reagent. IV. Determination of Zinc in presence of Copper, Silver and Mercury. P. R. Rây and T. C. Sarkar. (*Mikrochem.*, 1939, 27, 64-66.)—Zinc may be determined as quinaldinate in presence of copper by masking the reaction of the copper with thiourea, with which it forms a complex cation stable in acid solutions. Thiourea has been used for the determination of zinc as quinaldinate in presence of silver and mercury on the macro-scale (Ray and Dutt, *Z. anal. Chem.*, 1939, 115, 265; Abst., ANALYST, 1939, 64, 229), and may also be used on the micro-scale. For the determination of 0.15 to 1 mg. of zinc in presence of copper in aqueous solution, 0.3 to 0.5 ml. of a freshly prepared 20 per cent. sodium bisulphite solution is added to reduce any cupric copper to the cuprous state. This is followed by the dropwise addition of 0.05 ml. of acetic acid and 1-1.5 ml. of a 10 per cent. solution of thiourea. The beaker is heated on the water-bath, and the zinc is precipitated with an excess (0.2-1 ml.) of sodium quinaldinate solution (1 per cent. quinaldinic acid). After settling, the precipitate is filtered off, washed 5 or 6 times with 0.5 to 1 ml. of hot water, and dried in a current of air at 125° C. The same procedure is used in presence of mercury and silver, except that the sodium bisulphite may be omitted. When silver, mercury and copper are present together, a little potassium iodide must be added before the sodium bisulphite, but the separation is less satisfactory than from the individual elements. The Emich filter-stick procedure is used throughout. J. W. M.

Physical Methods, Apparatus, etc.

New Cell for Electrodialysis, especially of Soils. A. W. Marsden. (*J. Soc. Chem. Ind.*, 1940, 59, 60-62r.)—Electrodialysis is preferable to leaching methods for the determination of exchangeable bases in soils, because the bases are exchanged for hydrogen ions and appear as hydroxides in the cathode chamber, and there is no need to evaporate large volumes of solution and to remove the leaching agent before analysis. Existing cells are reviewed and criticised, and arguments are put forward in favour of a 3-chamber cell of the following design:—The central chamber is spherical in shape (to avoid "dead" spaces) and has a capacity of 30 ml. It is provided with a rotating stirrer which is inserted through a neck 7 cm. long in the top of the sphere, and on each side are interchangeable end-chambers which are separated from the sphere by means of small disc-shaped membranes of cellophane or other suitable material. The end-chambers are horizontal cylinders and have outlets, at the top to facilitate washing and the escape of electrolytic gases, and at the bottom to enable the liberated acids and bases to be withdrawn for analysis. The outer ends are closed with rubber stoppers, through which pass the glass tubes which hold the stout platinum wires supporting the disc-shaped vertical platinum electrodes (thickness 0.006 in., diameter 19 to 20 mm.). These electrodes are 4 cm. apart and each is drilled with 4 holes (diameter, 3 mm.) so as to facilitate the escape of gas bubbles, the accumulation of which would increase the internal resistance of the cell. Since the 3 chambers are held together by springs attached to the collars of the end-chambers, dismantling, cleaning and assembly are greatly facilitated. The procedure is to place 10 g. of air-dry soil in the central chamber, to fill the end-chambers with distilled water, and, after the current (D.C., 220 volts) has been switched on, to add 30 ml. of water to the central chamber and start the stirrer. As a rule the current does not rise above 60 to 100 milliamp. during electrodialysis, but should it do so, a 25-watt lamp may be included in the circuit. The temperature may be kept at 30° to 40° C. by changing the anode and cathode liquids hourly for the first few hours. This, however, may

also increase the resistance of the cell, although if advantage is taken of the fact that more base is liberated than acid and the cathode liquid only is changed, this effect is minimised. As a rule, 8 to 10 hours are required for complete electro-dialysis, and the current is then about 5 milliamp. The results obtained are satisfactorily reproducible, and the cell appears to be suitable for the examination of other colloidal substances. The high electro-osmotic flow to the cathode, which is typical of 2-chamber cells, necessitating also the evaporation of a large volume of dialysate before analysis, is avoided; the possibility of any action of the liberated acids on the soil is eliminated, and these liberated acids may be removed for analysis.

J. G.

Reviews

PRACTICAL PHARMACEUTICAL CHEMISTRY. By F. N. APPLEYARD, B.Sc., F.I.C., Ph.C., and C. G. LYONS, M.A., Ph.D., A.I.C. 4th Edition. Pp. 174 + vii. London: Sir Isaac Pitman & Sons. 1939. Price 6s. 6d.

The appearance of a fourth edition of this useful little manual is in itself a sufficient recommendation of its value in the training of pharmacists. The previous edition was made necessary by certain alterations in the syllabus for the Chemist and Druggist Qualifying Examination, for which the book is specially written, and in this edition the new material, chiefly concerned with qualitative organic analysis, has been extended.

Nearly half the book is devoted to exercises on volumetric analysis, some of which could possibly be omitted in a future edition without detracting from the book's usefulness. In addition, there are short chapters, presumably adequate for their purpose, on gravimetric analysis, official limit tests, alkaloidal assay processes and, rather unexpectedly, the preparation of organic compounds. All the exercises are carefully selected, both for their value as a means of learning chemistry and for their interest to the future pharmacist as being carried out on substances that are of importance pharmaceutically. The final chapter, on qualitative organic analysis, contains the most recent additions and is a bold attempt to compress a difficult subject into a small space. Again the examples given are substances of pharmaceutical importance.

The book is carefully written and the instructions are clear and precise. Indeed, the only fault that can be found with the book is that it is a little too precise. Its instructions do not allow of any initiative—or of any mistakes even—and are reminiscent of the official methods of assay in the British Pharmacopoeia, to which, in a way, the book serves as an introduction. One feels that the student may finish his course with the impression that he now knows all there is to know about analytical and organic chemistry, when in effect he has learned a little about the behaviour of a few of the more important compounds that have applications in pharmacy.

F. A. ROBINSON

ANNUAL REPORTS OF THE PROGRESS OF APPLIED CHEMISTRY. Vol. XXIV for 1939. Issued by the Society of Chemical Industry. Pp. 756. Price to members, 7s. 6d.; to non-members, 12s. 6d.

In praise of the current volume of this well-known annual series of reports it is sufficient to say that it has all the virtues accorded by reviewers to its predecessors. The reports are arranged under the now familiar headings, and each contains the detailed account of progress in the past year required by the specialist, skilfully presented so as to form a lucid survey intelligible to the general reader.

In some of the reports progress in analytical work in a particular field is made the subject of a special section, in others it is not separated from the general subject-matter. Although there are no recorded advances in analytical procedure so outstanding as to merit special mention, the analytical chemist will find much that is applicable, either directly or with modification, to his own problems.

There are indications that many of the reports were cast into their final form some three months after the outbreak of war, and numerous references to our national needs are scattered throughout the volume. The reader will find comforting assurances that our chemical industries are fully prepared to deal with the demands that are, and will be, placed upon them.

Reports such as these must of necessity contain much highly specialised technical language. This has been used judiciously and never leads to obscurity, although there are a few brief instances of its degeneration into the cacophonous jargon associated with some industrial operations. Of the very few typographical errors (the phrase "substitutes to petrol" on p. 56 is presumably one of these), the only serious one is the appearance of "cistine" instead of "cystine" in the subject index. In one report the phrase "substituted by" affords an example of the use of "substitute" as if it were synonymous with "replace"—an error occurring with irritating frequency in contemporary scientific literature. These, however, are minor faults in a volume that presents a comprehensive survey of the work of the past year in a concise, interesting and often entertaining manner.

A. O. JONES

THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

Deaths

WITH deep regret we record the deaths of the following members:

Hugh Charles Loudon Bloxam, on July 14th.

Christopher Rawson, on May 30th.

The Use of Thiocyanogen Values in the Determination of Linoleic and Linolenic Acids and their Glycerides

By T. P. HILDITCH, D.Sc., F.I.C., AND K. S. MURTI, B.A., Ph.D., A.I.C.

THE selective addition of thiocyanogen to linoleic and linolenic acids and their derivatives, in conjunction with their iodine values, was put forward by Kaufmann¹ in 1925 as a method for the determination of mixtures of oleic, linoleic and linolenic acids or their esters (including glycerides); it has since found considerable use in the analysis of fatty oils, especially in the hands of Kaufmann and his collaborators. Whereas halogens (*e.g.* iodine monochloride or monobromide) react completely and additively with each ethenoid linkage in oleic, linoleic or linolenic acids, thiocyanogen, according to Kaufmann, unites quantitatively with oleic acid, but with only one of the two ethenoid linkages in linoleic acid, and with two of the three ethenoid linkages in linolenic acid. Clearly, the validity of the thiocyanogen method of analysis depends upon two factors: (*a*) the degree of precision of the method—how far results are reproducible by the same or different analysts, and (*b*) the correctness of the fundamental assumption regarding the extent of the selective addition of thiocyanogen to linoleic and linolenic compounds.

Doubt has been expressed by some workers (van der Veen,² Gay,³ Smith and Chibnall⁴) as to the possibility of obtaining reproducible results when addition of thiocyanogen is used as a quantitative method of analysis; but others (*e.g.* Waterman and Bertram,⁵ Jamieson and Baughman,⁶ Griffiths and Hilditch⁷) have found that the procedure can be made satisfactory in this respect, provided that most careful precautions are observed in the manipulative technique, especially in ensuring complete absence of moisture from all the reagents and apparatus, and in observing rigidly standardised conditions of procedure at all stages of the analysis. Admittedly, however, it is difficult to obtain, in the determination of thiocyanogen values, the same degree of accordance which characterises that of ordinary iodine values, but the disparity is not sufficiently great to interfere seriously with the usefulness of the method. Nevertheless, the time of contact of the thiocyanogen

reagent with the fatty compounds under examination is an important factor, to which further reference must be made below.

The main object of the present communication is to point out that Kaufmann's fundamental assumptions as to the "theoretical" thiocyanogen values of linoleic and linolenic acid appear to be ill-founded; with linoleic acid, thiocyanogen reacts to the extent of somewhat more than half the total unsaturation, whilst it reacts with considerably less than the assumed two-thirds of the total unsaturation in the triethenoid linolenic acid. We have had reason to suspect this for some time, and began a systematic investigation of the point more than a year ago. Pressure of other work prevented us from devoting full attention to this problem, and in the meantime other authors have published two communications (*v. infra*) which accord for the most part with our own observations. Since, however, we have approached the matter on somewhat different lines from either of the authors in question, an account of our results may be a useful further contribution to the discussion. The use of thiocyanogen values is not indispensable in the analysis of fats which contain only oleic and linoleic acids, but it is the only method, at present proposed, which affords a possibility of accurately determining oleic, linoleic and linolenic acids when all three are simultaneously present. Unfortunately, our results indicate that the thiocyanogen value of linolenic acid differs so much from that hitherto assumed by Kaufmann that calculations based on his value must be regarded as very uncertain.

We decided to prepare specimens of oleic, linoleic and linolenic acids, and of their methyl and ethyl esters, of as high a degree of purity as possible, and to examine the thiocyanogen values of the individual acids and esters, and also of various binary or ternary mixtures made up from the acids (or from the corresponding esters) in known proportions.

In the determination of the thiocyanogen value, a time of contact of exactly 24 hours was used, in accordance with previous custom in this laboratory and with the procedure adopted by most workers. Recently Riemenschneider and Wheeler⁸ have advocated a contact time of three hours, on the grounds that after this period has elapsed the thiocyanogen value of methyl linoleate accords closely with Kaufmann's "theoretical" value (*i.e.* addition at exactly half of the total unsaturation), whilst that of methyl oleate is only fractionally below the theoretical value (*i.e.* 100 per cent. addition). These authors show that, after more than three hours' contact, methyl oleate undergoes a further very small increase in thiocyanogen value, finally reaching, but not exceeding, the value (85.8) corresponding with 100 per cent. addition of thiocyanogen; also, that the thiocyanogen value of methyl linoleate (having reached 50 per cent. addition of thiocyanogen in three hours) continues to increase, but more and more slowly, up to, and beyond, 24 hours. Our reasons for preferring the latter time of contact of the thiocyanogen reagent are as follows:

(i) Iodine values are based primarily on conditions of addition which show 100 per cent. addition of halogen to oleic acid or its esters, and it seems logical to adopt the same substance for calibration in thiocyanometric analysis. General experience, we believe, is in favour of the longer period of contact, to ensure 100 per cent. addition of thiocyanogen to oleic acid. (ii) Although addition of thiocyanogen to linoleic compounds has not ceased after 24 hours, the rate of addition at that period is much slower than at 3 hours (when it is still proceeding with moderate rapidity). Any error in timing would therefore have much greater effect after 3 hours' than after 24 hours' contact. (iii) The time factor in the addition of thiocyanogen to linolenic acid has not been explored in so much detail as for the other two acids, but the relatively low values observed in the case of linolenic acid and its esters (*v. infra*) suggest that addition is still incomplete even after 24 hours. (iv) In our experience the rates of addition of thiocyanogen to free fatty acids are liable to be slower than to their esters.

We have therefore continued to adhere, in the present work, to a time of contact of 24 hours in the thiocyanogen value determinations.

EXPERIMENTAL

PREPARATION OF THE PURE FATTY ACIDS AND ESTERS.—The oleic acid and methyl oleate used were derived from accumulated fractions of almost pure C_{18} unsaturated esters which were fortunately available from ester-fractionation analyses of the "liquid" or mainly unsaturated acids of a group of beef fats. The latter contain only small proportions of linoleic acid, so that the ester-fractions employed already consisted almost wholly of methyl oleate.

Oleic Acid.—The acids (69 g.) from a portion of the combined methyl oleate fractions were first crystallised from acetone (690 ml.) at -20°C . for 2 hours, after which a crystalline deposit (about 9 g.) was removed; this contained a certain amount of palmitic acid. The portion in solution was crystallised from 720 ml. of acetone at -60°C . for $3\frac{1}{2}$ hours, and the deposited crystals were recrystallised from acetone at -60°C . for 3 hours. The recrystallised oleic acid (36.7 g.; iod. val. 89.1) was then fractionally distilled in a vacuum through an electrically-heated and packed column, when a main fraction of 21 g. (iod. val. 89.7; CNS val. 89.0) was obtained. This material, which contained at most less than 1 per cent. each of palmitic and linoleic acids, was used in the tests described below.

Methyl Oleate.—A further portion (63 g.) of the combined methyl oleate fractions mentioned was crystallised from acetone (630 ml.) at -37°C . for 2 hours to remove traces of methyl palmitate as far as possible, the soluble fraction being further crystallised from 12 volumes of acetone at -60°C . for 3 hours. The deposited crystals (50.5 g.; iod. val. 84.8) were collected and fractionally distilled in a vacuum through the column mentioned, when three fractions of almost pure methyl oleate were obtained. The middle one of these fractions (12.0 g.; iod. val. 86.0; CNS val. 85.3) was employed in the present work.

Linoleic Acid and Methyl Linoleate.—These were prepared by debromination of pure crystalline tetrabromostearic acid, obtained from the unsaturated acids of cottonseed oil (which contains no linolenic acid). A large stock (340 g.) of the tetrabromostearic acid was prepared from refined cottonseed oil (800 g.) by brominating the unsaturated acids in light petroleum (b.p. $80-100^{\circ}\text{C}$.) and crystallising the product several times from light petroleum (b.p. $40-60^{\circ}\text{C}$.); it melted sharply at 115°C . (Br found 53.2; calc. 53.3 per cent.).

Linoleic Acid was prepared by debromination of the tetrabromostearic acid (40 g.) in pyridine solution with zinc by a modified form of Kaufmann and Mestern's method⁹; the acid (18.1 g.) so obtained was distilled in a vacuum, and the main fraction (15.5 g.; iod. val. 180.0) was used in the further experiments.

Methyl Linoleate was similarly prepared by Rollett's process¹⁰ by debromination of tetrabromostearic acid (40 g.) with zinc and hydrogen chloride in methyl alcohol. The crude ester (19.5 g.) yielded on vacuum distillation 17.0 g. of methyl linoleate (iod. val. 172.9).

Linolenic Acid and Ethyl Linolenate.—These were similarly obtained from hexabromostearic acid, which was prepared by addition of bromine to the mixed fatty acids of linseed oil in ethereal solution. After repeated boiling of the crude product with ether, the purified hexabromostearic acid (235 g., from 520 g. of refined linseed oil) melted at $180.5-181^{\circ}\text{C}$. (Br found 62.9; calc. 63.3 per cent.).

Linolenic Acid was prepared by debromination of the hexabromostearic acid (27 g.) with zinc and pyridine; the crude acid (9.7 g.) gave on vacuum distillation 7.7 g. of slightly yellow-coloured linolenic acid (iod. val., found 267.4; calc. 274.1).

Ethyl Linolenate was prepared by Rollett's method from the hexabromostearic acid (25 g.) with zinc, hydrogen chloride and ethyl alcohol (the hexabromostearic acid is insufficiently soluble in boiling methyl alcohol to permit the ready

preparation of the methyl ester). The crude product (9.5 g.) yielded on distillation in a vacuum a colourless specimen of ethyl linolenate (7.8 g.; iod. val., found 246.8; calc. 249.0).

IODINE AND THIOCYANOGEN VALUES OF THE PURE INDIVIDUAL COMPOUNDS.—The following precautions were taken in connection with the preparation and use of the thiocyanogen reagent:—(a) Lead thiocyanate (AnalaR) was dried for at least a week in an evacuated exsiccator over phosphorus pentoxide. (b) Glacial acetic acid was refluxed with 15 per cent. of acetic anhydride for 2–3 hours, cooled, and kept for a few days before use in preparing the reagent. (c) A suspension of the dried lead thiocyanate (5 per cent.) in the prepared acetic acid was "aged" for about a week before use in the preparation of the actual reagent. (d) All apparatus and filter-paper employed were dried immediately before use, first in a steam oven and then at about 120° C. for at least an hour in an electrically-heated oven. (e) The reagent was always freshly prepared, by addition of bromine to the solution (c), immediately before use in determination of a batch of thiocyanogen values. Its strength was maintained close to $N/5$, and each batch of reagent was filtered twice to ensure an absolutely clear solution. (f) The amounts of acid or ester were so adjusted that the excess of reagent (20 ml.) was in every instance between 150 and 200 per cent. (g) The reagent (20 ml.) was left in contact with the fat (0.1–0.2 g. dissolved in 20 ml. of the prepared acetic acid) for 24 hours in the dark; 20 ml. of 20 per cent. aqueous potassium iodide solution were then added immediately prior to titration with standard thiosulphate solution.

The iodine and thiocyanogen values of the individual acids and esters used in the subsequent tests are given in Table I, together with the theoretical iodine values and the thiocyanogen values calculated according to Kaufmann's assumptions (addition to one of the double bonds in oleic and linoleic acids, and to two of three double bonds in linolenic acid).

TABLE I

| Compound | Iodine value | | Thiocyanogen value | |
|--------------------|--------------|-------|--------------------|------------------|
| | Found | Calc. | Found | Calc. (Kaufmann) |
| Oleic acid | 89.8 | 90.1 | 89.0 | 90.1 |
| Methyl oleate | 86.0 | 85.8 | 85.3 | 85.8 |
| Linoleic acid | 180.0 | 181.4 | 95.9 | 90.7 |
| Methyl linoleate | 172.9 | 172.8 | 91.8 | 86.4 |
| Linolenic acid | 267.4 | 274.1 | 162.5 | 182.7 |
| Ethyl linolenate | 246.8 | 249.0 | 146.6 | 166.0 |

IODINE AND THIOCYANOGEN VALUES OF BINARY AND TERNARY MIXTURES OF THE ACIDS OR ESTERS.—We proceed to report the rest of our experimental data before considering further the deviation between the observed thiocyanogen values of linoleic and linolenic acids and their esters and the values as calculated on Kaufmann's assumption. Five binary and three ternary mixtures of the acids, and also of the corresponding esters, were made up in the following proportions by weight:

| Acids | | | | Esters | | | |
|----------|-------|----------|-----------|----------|---------------|------------------|------------------|
| Ref. No. | Oleic | Linoleic | Linolenic | Ref. No. | Methyl oleate | Methyl linoleate | Ethyl linolenate |
| A1 | 74.5 | 25.5 | — | E1 | 76.0 | 24.0 | — |
| A2 | 51.5 | 48.5 | — | E2 | 48.0 | 52.0 | — |
| A3 | 25.1 | 74.9 | — | E3 | 33.0 | 67.0 | — |
| A4 | 50.2 | — | 49.8 | E4 | 49.8 | — | 50.2 |
| A5 | — | 49.3 | 50.7 | E5 | — | 50.3 | 49.7 |
| A6 | 31.3 | 35.7 | 33.0 | E6 | 32.4 | 35.7 | 31.9 |
| A7 | 48.4 | 25.2 | 26.4 | E7 | 48.2 | 26.7 | 25.1 |
| A8 | 24.3 | 50.2 | 25.5 | E8 | 24.1 | 50.5 | 25.4 |

The iodine values (Wijs) and thiocyanogen values of these mixtures are given in Table II, together with the values calculated numerically from the observed data in Table I. To illustrate the degree of reproducibility obtained, the individual determinations made are recorded, with the final mean value adopted; figures in brackets were not used for the calculation of the mean values.

TABLE II
OBSERVED IODINE AND THIOCYANOGEN VALUES OF THE MIXTURES OF
ACIDS OR ESTERS

| (i) <i>Iodine Values</i> | | | | | | | |
|--------------------------|--------------|-------|-------|----------|--------------|-------|-------|
| Acids | | | | Esters | | | |
| Ref. No. | | Mean | Calc. | Ref. No. | | Mean | Calc. |
| A1 | 112.6, 112.8 | 112.7 | 112.8 | E1 | 107.1, 106.7 | 106.9 | 106.7 |
| A2 | 133.1, 133.3 | 133.2 | 133.5 | E2 | 130.7, 130.7 | 130.7 | 131.1 |
| A3 | 157.0, 156.9 | 157.0 | 157.3 | E3 | 143.4, 143.3 | 143.4 | 144.1 |
| A4 | 177.6, 177.1 | 177.4 | 178.5 | E4 | 166.0, 166.6 | 166.3 | 166.8 |
| A5 | 224.0, 224.7 | 224.4 | 224.3 | E5 | 209.4, 209.1 | 209.3 | 209.6 |
| A6 | 180.7, 180.7 | 180.7 | 180.6 | E6 | 168.5, 168.5 | 168.5 | 168.2 |
| A7 | 159.6, 159.4 | 159.5 | 159.4 | E7 | 149.7, 149.6 | 149.7 | 149.6 |
| A8 | 180.2, 180.6 | 180.4 | 180.3 | E8 | 170.7, 170.5 | 170.6 | 170.7 |

| (ii) <i>Thiocyanogen Values</i> | | | | | | | |
|---------------------------------|----------------------------|-------|-------|--|--|--|--|
| Acids | | | | | | | |
| Ref. No. | | Mean | Calc. | | | | |
| A1 | (89.2), 90.1, (88.7), 90.7 | 90.4 | 90.7 | | | | |
| A2 | 91.5, 90.8, (90.4) | 91.2 | 92.3 | | | | |
| A3 | 92.8, 93.7, (91.9) | 93.3 | 94.2 | | | | |
| A4 | (123.6), 122.3, 122.1 | 122.2 | 125.6 | | | | |
| A5 | 127.2, 127.8, (125.7) | 127.5 | 129.7 | | | | |
| A6 | (113.8), 112.8, 112.6 | 112.7 | 115.7 | | | | |
| A7 | 108.7, 108.5, (106.9) | 108.6 | 110.1 | | | | |
| A8 | 109.1, 109.2, (108.0) | 109.2 | 111.2 | | | | |

| Esters | | | | | | | |
|--------|-----------------------|-------|-------|--|--|--|--|
| E1 | 86.6, 86.7 | 86.7 | 86.8 | | | | |
| E2 | 88.8, 88.5 | 88.7 | 88.7 | | | | |
| E3 | 90.1, 89.4 | 89.8 | 89.7 | | | | |
| E4 | 117.1, 117.0 | 117.1 | 116.1 | | | | |
| E5 | 120.8, 119.8, (116.6) | 120.3 | 119.1 | | | | |
| E6 | 107.7, 107.1 | 107.4 | 107.2 | | | | |
| E7 | 102.7, 103.0 | 102.8 | 102.5 | | | | |
| E8 | 104.5, 104.6 | 104.6 | 104.2 | | | | |

It will be seen from Table II that the observed iodine values show good reproducibility and also accordance with the calculated figures, but that the thiocyanogen data are not so accordant in either respect. The agreement between the individual determinations is not altogether satisfactory, and the mean values are sometimes more than one unit divergent from the calculated figures for the mixtures. This is more noticeable in the acid mixtures than in the ester mixtures, both reproducibility and accordance with the calculated figures being fairly satisfactory in the latter. Further, the discrepancies only become marked when linolenic acid or ester is a component of the mixture. It may also be remarked here that we have encountered similar difficulties with different preparations of the individual linoleic and linolenic compounds; preparations, made at an early stage of the work, which had practically the same iodine values as those recorded in Table I, occasionally gave thiocyanogen values from 3 to 5 units lower than those which we have finally accepted from our later work.

It remains to compare the percentage proportions of the components of the various mixtures which we have examined as determined (a) from the observed thiocyanogen values of the individual components recorded in Table I, and (b) from the equations based on the original assumptions of Kaufmann. These figures are given in Table III.

TABLE III

PERCENTAGE PROPORTIONS OF COMPONENTS OF THE MIXTURES

(a) Calculated from the observed thiocyanogen values in Table I.

(b) Calculated from the theoretical assumptions of Kaufmann.

| Ref. No. | Oleic | | | Linoleic | | | Linolenic | | |
|---------------|--------|--------------|--------------|----------|--------------|--------------|-----------|--------------|--------------|
| | Actual | Found (a) | Found (b) | Actual | Found (a) | Found (b) | Actual | Found (a) | Found (b) |
| <i>Acids</i> | | | | | | | | | |
| A1 | 74.5 | 73.5 | 75.4 | 25.5 | 26.5 | 24.6 | — | — | — |
| A2 | 51.5 | 50.1 | 53.7 | 48.5 | 49.9 | 46.3 | — | — | — |
| A3 | 25.1 | 24.3 | 29.8 | 74.9 | 75.7 | 70.2 | — | — | — |
| A4 | 50.2 | 47.4 | 39.6 | — | — | — | 49.8 | 52.6 | 60.4 |
| A5 | — | — | — | 49.3 | 60.3 | 66.5 | 50.7 | 39.7 | 33.5 |
| A6 | 31.3 | 25.9 | 25.2 | 35.7 | 46.5 | 50.7 | 33.0 | 27.5 | 24.1 |
| A7 | 48.4 | 45.2 | 44.1 | 25.2 | 31.8 | 36.2 | 26.4 | 23.0 | 19.7 |
| A8 | 24.3 | 20.8 | 21.8 | 50.2 | 57.4 | 58.0 | 25.5 | 21.8 | 20.2 |
| <i>Esters</i> | | | | | | | | | |
| E1 | 76.0 | 75.1 | 76.6 | 24.0 | 24.9 | 23.4 | — | — | — |
| E2 | 48.0 | 48.2 | 51.4 | 52.0 | 51.8 | 48.6 | — | — | — |
| E3 | 33.0 | 33.9 | 38.0 | 67.0 | 66.1 | 62.0 | — | — | — |
| E4 | 49.8 | 50.9 | 40.7 | — | — | — | 50.2 | 49.1 | 50.3 |
| E5 | — | — | — | 50.3 | 41.4 | 62.3 | 49.7 | 58.6 | 37.7 |
| E6 | 32.4 | 32.2 | 28.2 | 35.7 | 35.8 | 45.2 | 31.9 | 32.0 | 26.6 |
| E7 | 48.2 | 48.3 | 45.0 | 26.7 | 26.2 | 33.9 | 25.1 | 25.5 | 21.1 |
| E8 | 24.1 | 24.8 | 22.7 | 50.5 | 49.2 | 54.2 | 25.4 | 26.0 | 23.1 |

DISCUSSION.—The data in Table III point to two main conclusions:

(i) Calculations based on the theoretical assumptions of addition of thiocyanogen to exactly one double bond in linoleic compounds and to exactly two double bonds in linolenic compounds lead to results which, when the proportion of linoleic or linolenic compounds in the mixtures is at all considerable, are widely at variance with the actual proportions present; whereas employment of the actual thiocyanogen values observed for the individual compounds (Table I) gives figures which in general accord closely with the actual facts.

(ii) Calculations on the latter basis, however, in some instances (A5, A6, A7, A8, E5) fail to give figures in accordance with the facts. These are, of course, the instances in which unsatisfactory agreement was noted in Table II between the observed and calculated thiocyanogen values of the mixtures. As already stated, the agreement is in general satisfactory for the ester-mixtures, although even here the method appears to begin to give erratic results when the proportion of polyethenoid material is high, or when the content of linolenic acid exceeds about 25 per cent.; or, put in another way, when the iodine value of the mixture exceeds 170.

The information in Table III establishes, however, that (apart from the instances noted in the previous paragraph) the empirically determined thiocyanogen values of the pure compounds are operative in the mixtures, and should therefore be accepted in lieu of the formerly adopted "theoretical" thiocyanogen values for linoleic and linolenic compounds. It seems unfortunate, indeed, that so arbitrary an assumption as *exact* addition at one or at two double bonds, as the case may be, should have been accepted, apparently without adequate experimental verification. Kaufmann¹ observed a thiocyanogen value of 82.5 for a glyceride

"trilinolein" of iodine value 169.1, but seems not to have recorded a thiocyanogen value for pure linolenic acid (although Kaufmann and Keller¹ (p. 74) mention a specimen of linolenic acid prepared by Rollett's method¹⁰ with an iodine value of 268.0). Other workers have recorded for individual compounds values which are not very dissimilar from our own, as follows (Table IV):

TABLE IV
THIOCYANOGEN VALUES RECORDED FOR LINOLEIC ACID, METHYL
LINOLEATE AND LINOLENIC ACID

| Observers | Linoleic acid | Methyl linoleate | Linolenic acid |
|--|---------------|------------------|----------------|
| Kimura ¹¹ | — | 89.9 | — |
| Waterman, Bertram and van Westen ¹² | 92.9 | — | — |
| Brown and Shinowara ¹³ | — | — | 161.0 |
| Riemenschneider and Wheeler ⁴ | — | 89.0 | — |
| Kass, Lundberg and Burr ¹⁴ | 96.3 | — | 171.0 |
| Present work | 95.9 | 91.8 | 162.5 |

The figures in Table IV all refer to compounds prepared by debromination of crystalline bromo-adducts of the respective unsaturated acids, and it is possible to argue that the thiocyanogen values of the natural acids may be different; although it is now generally accepted that the product of debromination of the crystalline tetrabromostearic acid (formerly termed " α -linoleic acid") is identical with the natural product (Brown and Frankel,¹⁵ Riemenschneider, Wheeler and Sando,¹⁶ Hilditch and Jasperson¹⁷). There is, however, evidence in the studies of Brown and his co-workers, who have prepared highly concentrated forms of linoleic and linolenic acids by processes involving only the use of crystallisation from solvents at low temperatures, that the linolenic acid of seed fats has the same thiocyanogen value as that of the acid regenerated from crystalline hexabromostearic acid. Brown and Shinowara¹³ have recorded the iodine and thiocyanogen values of four highly concentrated preparations of natural linolenic acid, which they obtained by repeated low-temperature crystallisation from solvents of the fatty acids from linseed or perilla oils (Table V). It may be taken for granted that the accompanying impurity in these specimens will be confined to linoleic acid (also present in the seed fats used), and in this instance the percentages of linolenic acid present can be determined from the iodine values, and then the thiocyanogen value of the linolenic acid can be calculated after allowance for that of the linoleic acid present. We have applied these calculations to Brown and Shinowara's data for the four concentrates of linolenic acid, employing for the thiocyanogen value of linoleic acid both the value observed by us in the present work and also the previously assumed "theoretical" value of 90.7 (Table V).

TABLE V
DEDUCED THIOCYANOGEN VALUES OF LINOLENIC ACID ISOLATED BY LOW-
TEMPERATURE CRYSTALLISATION (BROWN AND SHINOWARA¹³)

| Concentrate | Iodine value | CNS value | Linolenic acid present | | |
|-------------|--------------|-----------|------------------------|---------------|---------------|
| | | | Per Cent. (calc.) | CNS value (a) | CNS value (b) |
| 1 | 258.1 | 150.1 | 82.7 | 161.5 | 162.5 |
| 2 | 260.0 | 152.6 | 84.8 | 162.8 | 163.8 |
| 3 | 262.0 | 154.1 | 86.9 | 162.9 | 163.7 |
| 4 | 259.0 | 151.3 | 83.7 | 162.1 | 163.1 |
| | | | (Mean:) | 162.3 | 163.3) |

(a) Linoleic acid CNS value taken as 95.9.

(b) " " " " " " 90.7.

The calculated thiocyanogen value for natural linolenic acid is thus very close to that observed for the regenerated " α -linolenic acid" by Brown and Shinowara¹⁸ (161.0) and by ourselves (162.5); Kimura¹¹ recorded that methyl linolenate (iod. val., found 258.6; calc. 261.9) showed a thiocyanogen value of 152.0, corresponding with 159.6 for the acid, whilst our value for ethyl linolenate (146.6) corresponds with 161.4 for the acid. It is evident from all these data that the actual thiocyanogen value of linolenic acid, natural or regenerated, is close to 162 and much lower than the figure of 182.7 which has hitherto been employed.

TABLE VI

ALTERATIONS IN OLEIC, LINOLEIC AND LINOLENIC ACID PERCENTAGES OF CERTAIN FATS BY THE ADOPTION OF THE REVISED THIOCYANOGEN VALUES FOR LINOLEIC AND LINOLENIC ACIDS

| Fat | Observers | Fatty acid percentages calculated with CNS values | | | | | |
|------------------|---|---|-------------------------------|--------------------------------|----------------------------------|-------------------------------|--------------------------------|
| | | Linoleic 90.7 Linolenic 182.7 | | | Linoleic 95.9 Linolenic 162.5 | | |
| | | Oleic Per Cent. | Lin- oleic Per Cent. | Lino- lenic Per Cent. | Oleic Per Cent. | Lin- oleic Per Cent. | Lino- lenic Per Cent. |
| Linseed— | | | | | | | |
| Calcutta | Kaufmann and Keller ¹ | 12.5 | 34.1 | 42.1 | 14.3 | 29.2 | 45.2 |
| La Plata | " " | 8.0 | 46.7 | 36.3 | 16.1 | 30.9 | 44.0 |
| " " | Griffiths and Hilditch ⁷ | 9.6 | 42.6 | 38.1 | 18.8 | 24.2 | 47.3 |
| Perilla | Kaufmann ¹⁸ | 3.9 | 44.3 | 44.2 | 13.7 | 16.0 | 62.7 |
| " " | " | 10.7 | 33.6 | 49.0 | 23.0 | 0.1 | 70.2 |
| Stillingia seed— | | | | | | | |
| Chinese | Jamieson and McKinney ¹⁹ | 11.1 | 52.8 | 26.8 | 15.8 | 45.5 | 29.4 |
| American | " " | 8.1 | 59.4 | 25.9 | 11.9 | 53.1 | 28.4 |
| Candlenut | Jamieson and McKinney ²⁰ | 27.7 | 41.7 | 21.7 | 29.4 | 40.0 | 21.7 |
| Wallflower seed | van Loon ²¹ | 5.2 | 26.1 | 20.5 | 7.7 | 21.3 | 22.8 |
| " " | Griffiths and Hilditch ⁷ | 8.1 | 35.2 | 14.2 | 10.0 | 31.5 | 16.0 |
| Hempseed | Kaufmann and Juschkevitch ²² | 12.6 | 53.0 | 24.3 | 16.0 | 45.8 | 28.1 |
| " " | Griffiths and Hilditch ⁷ | 6.7 | 68.8 | 15.0 | 6.2 | 69.9 | 15.3 |
| Walnut | Jamieson and McKinney ²³ | 36.4 | 50.0 | 7.7 | 34.7 | 56.7 | 2.7 |
| " " | Ueno and Nishikawa ²⁴ | 12.2 | 69.3 | 7.7 | 8.8 | 76.0 | 4.4 |
| " " | Griffiths and Hilditch ⁷ | 19.1 | 65.9 | 6.9 | 15.6 | 72.6 | 3.6 |
| Soya-bean | " " | 26.1 | 54.7 | 5.8 | 23.3 | 60.3 | 3.0 |
| " " | Hilditch and Jaspersen ²⁵ | 28.5 | 51.3 | 5.6 | 26.9 | 58.0 | 2.0 |
| Grape-seed | Jamieson and McKinney ²⁶ | 33.8 | 54.6 | 2.4 | 30.8 | 60.0 | — |

Kass, Lundberg and Burr¹⁴ have recently published work on similar lines to the present investigation in which they reach the conclusions that "the true thiocyanogen value of linoleic acid and the optimum conditions for determining it call for further consideration," and that "there can be little doubt that the thiocyanogen value of linoleic acid is an empirical value . . . which is appreciably higher than required by theory for half-saturation." They state, pending the result of further work*, that a preliminary investigation of a linolenic acid with iodine value 270.5 gave thiocyanogen values of 171 (24 hours' contact) and 175.6 (48 hours' contact); these figures, however, are higher than any of those to which we have just made reference. Kass *et al.* suggest the substitution of the empirical values for the hitherto accepted ones in the calculation of the proportions of unsaturated acids in fats.

* In a subsequent paper (*Oil and Soap*, 1940, 17, 118), published while the present communication was in the press, Kass, Loeb, Norris and Burr report the thiocyanogen value of " α -linoleic acid" to be 167.3, and formulate conclusions in close general agreement with those here put forward.

In the light of what has been pointed out in the present discussion, we are emphatically of the same opinion, provided that satisfactory agreement can be reached as to the true empirical thiocyanogen values of the two acids under accepted and rigidly standardised conditions of determination. The comparatively small difference between the observed and hitherto accepted thiocyanogen values of linoleic acid has a correspondingly small and perhaps not too serious effect on the calculated proportions of that acid in fats from which linolenic acid is absent (or present only in very small amounts). But the matter is widely different with fats of which linolenic acid is a major component—for which fats, as we have already pointed out, thiocyanometric analysis is most necessary and valuable. Here, the substantial difference in the values adopted for linolenic acid frequently has the result of altering in the same direction by several units per cent. both the percentages of oleic and linolenic acid, with a corresponding doubled effect in the opposite direction on the percentage of linoleic acid. In illustration of this point, we have collected in Table VI the results of typical analyses, as given by various workers, on some of the more unsaturated fatty oils in terms of the hitherto accepted thiocyanogen values of linoleic (90.7) and linolenic (182.7) acids, and have added thereto revised calculations based on our observed values of, respectively, 95.9 and 162.5. This table is for the time being merely illustrative; we consider that general agreement as to the values to be adopted must precede any final authoritative revision of previously published data.

SUMMARY.—The iodine and thiocyanogen values of carefully purified oleic, linoleic and linolenic acids, methyl oleate, methyl linoleate and ethyl linolenate have been determined. The thiocyanogen values (0.2 *N* reagent, 24 hours' contact, 150–200 per cent. excess) of linoleic and linolenic acids are respectively 95.9 and 162.5, corresponding values being observed for the respective esters. Both values differ from those originally postulated by Kaufmann¹ (respectively 90.7 and 182.7); the present results, especially for linolenic acid, accord well with recent data published by other workers.

Examination of binary and ternary mixtures of the pure acids or the pure esters shows that the observed values hold for the examination of mixtures of the compounds, subject to possible discrepancies when the unsaturation is high (iodine value above 170), especially with mixtures of the three acids.

It is recommended that agreed empirically determined values obtained under carefully prescribed conditions should replace the hitherto-accepted "theoretical" thiocyanogen values of linoleic and linolenic compounds; but that extensive revision of already published data should be deferred until general agreement has been reached as to the precise values and the conditions of determination.

Meantime, data obtained by thiocyanometric analysis for the component acids of fats which contain high proportions of linolenic and linoleic acids must be regarded as uncertain. Instances of the variations involved are quoted for a number of the more important liquid fats.

REFERENCES

1. H. P. Kaufmann, *Arch. Pharm.*, 1925, **263**, 675; *Abst.*, *ANALYST*, 1926, **51**, 157. *Z. Unters. Lebensm.*, 1926, **51**, 15; *Abst.*, *ANALYST*, 1926, **51**, 264. H. P. Kaufmann and M. Keller, *Z. angew. Chem.*, 1929, **42**, 20, 73; *Abst.*, *ANALYST*, 1929, **54**, 304.
2. H. van der Veen, *Chem. Umschau*, 1931, **38**, 117, 277.
3. P. J. Gay, *J. Soc. Chem. Ind.*, 1932, **51**, 126t.
4. J. A. B. Smith and A. C. Chibnall, *Biochem. J.*, 1932, **26**, 218.
5. H. I. Waterman and S. H. Bertram, *J. Soc. Chem. Ind.*, 1929, **48**, 79t.
6. G. S. Jamieson and W. F. Baughman, *Oil and Fat Ind.*, 1930, **7**, 419, 437.
7. H. N. Griffiths and T. P. Hilditch, *J. Soc. Chem. Ind.*, 1934, **53**, 75t.
8. R. W. Riemenschneider and D. H. Wheeler, *Oil and Soap*, 1939, **16**, 219.
9. H. P. Kaufmann and H. E. Mestern, *Ber.*, 1936, **69B**, 2684.
10. A. Rollett, *Z. physiol. Chem.*, 1909, **62**, 410.
11. W. Kimura, *J. Soc. Chem. Ind. Japan*, 1929, **32**, 141b.

12. H. I. Waterman, S. H. Bertram and H. A. van Westen, *J. Soc. Chem. Ind.*, 1929, **48**, 501; Abst., *ANALYST*, 1929, **54**, 252.
13. G. Y. Shinowara and J. B. Brown, *J. Amer. Chem. Soc.*, 1938, **60**, 2784.
14. J. P. Kass, W. O. Lundberg and G. O. Burr, *Oil and Soap*, 1940, **17**, 50.
15. J. B. Brown and J. Frankel, *J. Amer. Chem. Soc.*, 1938, **60**, 54; Abst., *ANALYST*, 1938, **63**, 211.
16. R. W. Riemenschneider, D. H. Wheeler and C. E. Sando, *J. Biol. Chem.*, 1939, **127**, 391.
17. T. P. Hilditch and H. Jasperson, *J. Soc. Chem. Ind.*, 1939, **58**, 233.
18. H. P. Kaufmann, *Allgem. Oel- u. Fett-Ztg.*, 1930, **27**, 39.
19. G. S. Jamieson and R. S. McKinney, *Oil and Soap*, 1938, **15**, 295.
20. ——— *id.*, 1937, **14**, 203.
21. J. van Loon, *Rec. trav. chim.*, 1930, **49**, 745.
22. H. P. Kaufmann and S. Juschkevitsch, *Z. angew. Chem.*, 1930, **43**, 90.
23. G. S. Jamieson and R. S. McKinney, *Oil and Soap*, 1936, **13**, 202.
24. S. Ueno and Y. Nishikawa, *J. Soc. Chem. Ind. Japan*, 1937, **40**, 313b.
25. T. P. Hilditch and H. Jasperson, *J. Soc. Chem. Ind.*, 1938, **57**, 84; Abst., *ANALYST*, 1938, **63**, 442.
26. G. S. Jamieson and R. S. McKinney, *Oil and Soap*, 1935, **12**, 241.

DEPARTMENT OF INDUSTRIAL CHEMISTRY
THE UNIVERSITY, LIVERPOOL

May 23rd, 1940

The Determination of Aluminium, Magnesium or Beryllium in Nickel Alloys

(Communication from the Staff of the Research Laboratories of The General Electric
Company, Limited, Wembley, England)

By R. C. CHIRNSIDE, F.I.C., L. A. DAUNCEY, B.Sc., AND P. M. C. PROFFITT

(Read at the Meeting, April 3rd, 1940)

It is the purpose of this paper to describe the results of an investigation into the most suitable methods of analysis for certain alloys used in the manufacture of radio valves and other thermionic devices. Nickel is the metal most commonly used for the electrodes, and it is well known that impurities in the nickel have a considerable effect on the thermionic properties. The addition of certain reducing elements, for example, has been found beneficial and an alloy containing 2 per cent. of aluminium has been employed.

A note on a method for the determination of the aluminium has previously been published by one of us.¹ Briefly, that method consisted in pouring a solution of the nickel alloy, to which potassium cyanide had been added in excess to form the soluble nickelocyanide, into a dilute ammonia solution, the precipitated aluminium hydroxide being filtered off, ignited and weighed.

The method proved generally satisfactory, and it was later adapted, with suitable modification, to the determination of magnesium in nickel-magnesium alloys of which the magnesium-content was of the order of 0.3 per cent. However, difficulties were sometimes encountered in the analysis of the aluminium alloys, and doubts arose as to the accuracy of the results obtained. Continued experience of the method, particularly with alloys containing only 0.4 per cent. of aluminium, confirmed the doubts as to its accuracy. One serious difficulty was the occasional co-precipitation, with the aluminium hydroxide, of a flocculent brown organic nickel compound, but erratic results were sometimes obtained even in absence of the brown precipitate.

The factors that were thought to contribute to the variability of the results included the formation of alkali by hydrolysis of the cyanide with consequent solution of aluminium hydroxide, the time of standing before filtration, and the temperature of the solution. The greater variability of the results obtained on

the alloy containing 0.4 per cent. of aluminium was consistent with the relatively smaller amount of aluminium hydroxide involved in this instance. Several synthetic solutions were prepared to represent alloys containing 0.4 per cent. and 2 per cent. of aluminium, and the aluminium-content was determined by the cyanide method embodying various modifications designed to explore the effects of the various factors mentioned above. Some of the results obtained are given in Table I, and it will be seen that none of the variations made in the method resulted in an accurate determination of the aluminium-content of a solution representing the 0.4 per cent. alloy. The experiments are not described in detail, as the method was subsequently abandoned.

In further experiments on synthetic solutions the aluminium was precipitated directly with ammonia and ammonium chloride. A considerable excess of ammonia was required to form the soluble nickel complex, and this entailed a risk of losing aluminium hydroxide by solution. On the other hand, the precipitate was invariably contaminated with nickel, so that the accuracy of the final result depended on a fortuitous compensation of errors. Some results obtained by the use of this method are included in Table I.

TABLE I
ALUMINIUM IN SYNTHETIC SOLUTIONS

| No. | Nickel added g. | Aluminium added | | Aluminium found | | Remarks |
|---------------------------|--------------------|-----------------|-----------|-----------------|-----------|--|
| | | mg. | per cent. | mg. | per cent. | |
| <i>Cyanide method</i> | | | | | | |
| 1 | 2.0 | 8.26 | 0.41 | 10.8 | 0.54 | |
| 2 | 2.0 | 7.96 | 0.40 | 5.5 | 0.28 | |
| 3 | 2.0 | 8.86 | 0.44 | 11.0 | 0.55 | |
| 4 | 2.0 | 8.63 | 0.43 | 10.5 | 0.53 | |
| 5 | 2.0 | 8.52 | 0.43 | 14.3 | 0.72 | |
| 6 | 2.0 | 40.0 | 2.00 | 42.6 | 2.13 | |
| 7 | 2.0 | 40.0 | 2.00 | 41.4 | 2.07 | |
| <i>Pptn. with ammonia</i> | | | | | | |
| 8 | 2.0 | 8.48 | 0.42 | 11.4 | 0.57 | One pptn. |
| 9 | 2.0 | 8.70 | 0.44 | 8.8 | 0.44 | One pptn. |
| 10 | 2.0 | 8.52 | 0.43 | 8.5 | 0.43 | Three pptns. Ppt. contained 0.05 mg. of NiO. |
| 11 | 2.0 | 8.66 | 0.43 | 9.7 | 0.49 | Three pptns. Ppt. contained 0.5 mg. of NiO. |

Further experiments were carried out on samples of nickel-aluminium alloy with nominal 0.4 per cent. and 2.0 per cent. aluminium-contents. The aluminium was determined by the cyanide method, by precipitation with barium carbonate, by precipitation with sodium succinate, and by precipitation with ammonia after removal of the nickel by electrolysis. The last-named method proved to be most suitable and has been adopted for our work, but for purposes of comparison the results obtained by all the methods mentioned are given in Table II.

TABLE II
ALUMINIUM IN NICKEL-ALUMINIUM ALLOYS
Aluminium found, per cent.

| No. | Cyanide | Barium carbonate | Sodium succinate | Electrolysis |
|-----|------------------------|------------------|------------------|--------------|
| 63 | 0.31, 0.27, 0.40 | 0.34 | — | — |
| 66 | 0.34, 0.38, 0.41, 0.41 | 0.45 | — | 0.40, 0.42 |
| 67 | 0.32, 0.32, 0.37, 0.41 | 0.35 | — | 0.34, 0.33 |
| | 0.34, 0.31, 0.72, 0.39 | | | |
| | 0.30, 0.40 | | | |
| 86 | — | 0.42, 0.38 | 0.38 | 0.35, 0.35 |
| 88 | — | — | 0.37, 0.38, 0.38 | 0.37, 0.37 |
| 87 | — | — | — | 1.91, 1.92 |

The results obtained with the electrolytic method were so encouraging that it was decided to examine the method in more detail, and for this purpose further experiments were carried out with synthetic solutions.

ELECTROLYSIS WITH A MERCURY CATHODE.—(i) *Preliminary Experiments on Synthetic Solutions.*—Fifty g. of Mond nickel pellets were dissolved in 400 ml. of nitric acid (1 : 1). The solution was filtered and made up to 500 ml., so that 20 ml. contained the equivalent of 2 g. of nickel. Weighed amounts of pure aluminium ribbon, to represent the 0.4 per cent. and 2.0 per cent. alloys, were dissolved in freshly-prepared sodium hydroxide solution and added to 20-ml. portions of the nickel solution.

The mixtures were evaporated to fuming with sulphuric acid, and the electrolysis and subsequent precipitation of the aluminium with ammonia were carried out as described under the heading "experimental procedure." A blank determination on 20 ml. of nickel solution yielded 0.6 mg. This was most probably derived from the reagents, glassware and filter-paper (*cf.* Etheridge, *ANALYST*, 1929, 54, 142). The results, corrected for the blank, are given in Table III.

TABLE III

| No. | Nickel added g. | Aluminium added | | Aluminium found | |
|-----|--------------------|-----------------|-----------|-----------------|-----------|
| | | mg. | per cent. | mg. | per cent. |
| 1 | 2.0 | 8.7 | 0.44 | 8.8 | 0.44 |
| 2 | 2.0 | 8.8 | 0.44 | 8.8 | 0.44 |
| 3 | 2.0 | 40.5 | 2.03 | 40.6 | 2.03 |
| 4 | 2.0 | 39.5 | 1.98 | 39.7 | 1.98 |

These experiments showed that the results obtained by the electrolysis of alloys and recorded in Table II were accurate as well as precise. It was therefore decided to develop a method for the routine determination of aluminium in nickel alloys by means of electrolysis over a mercury cathode.

(ii) *Development of the Method.*—Wolcott Gibbs² in 1883 first suggested the use of mercury as a negative electrode in electrolysis, and E. F. Smith³ later suggested many applications of the process. Cain,⁴ Lundell, Hoffman and Bright,⁵ and Etheridge⁶ have employed the method for the separation of vanadium from steel, and Brophy⁷ and Etheridge⁸ have used the method for the separation of aluminium from nickel-chromium alloys and steel respectively. B. S. Evans⁹ has described an apparatus employing a moving mercury cathode, designed to overcome most of the disadvantages associated with this method of analysis.

In the method as originally conceived by Gibbs the metals deposited in the mercury were determined, but so many elements may be deposited that the method came to be used chiefly for the removal of interfering elements from solution. Lundell and Hoffman¹⁰ give a table showing the elements quantitatively deposited in the mercury cathode, those which are quantitatively separated from the electrolyte but are not quantitatively deposited in the mercury, and those which are incompletely separated. Of the commoner elements, chromium, iron, cobalt, nickel, copper, zinc, cadmium, silver, tin and bismuth can be quantitatively separated from aluminium, magnesium, beryllium, titanium, zirconium and vanadium.

Manganese is said to be incompletely deposited in the mercury and on the anode. That this valuable and elegant method has found comparatively little use in analysis is due, in our opinion, to the belief that complicated and expensive apparatus is necessary. In devising an apparatus for our particular problem the main consideration were that it should be as simple as possible, and that a number of cells could be used at one time. In particular, the minimum number of parts was to be exclusive to this test, and special electrolysis stands, vessels and stirring devices were to be avoided if possible.

An electrode arrangement was constructed as in Fig. 1. A platinum anode was sealed into a glass tube, connection being made to it by means of a wire dipping into a small amount of mercury in the bottom of the tube. A short loop of stout platinum wire was sealed into a somewhat longer glass tube and a similar connection made to it. The two tubes were fastened by means of an elastic band to a grooved separator formed from a cork. The assembly was stood in a 250-ml. beaker, so that the loop rested on the bottom and near the side, and the lowest part of the anode was about 2 cm. from the bottom. The loop, which formed the connection to the cathode, was so shaped that it could be covered completely by 10 ml. of mercury in the bottom of the beaker. When the tubes rested in the lip of the beaker an ordinary watch-glass provided adequate protection against loss by spraying.

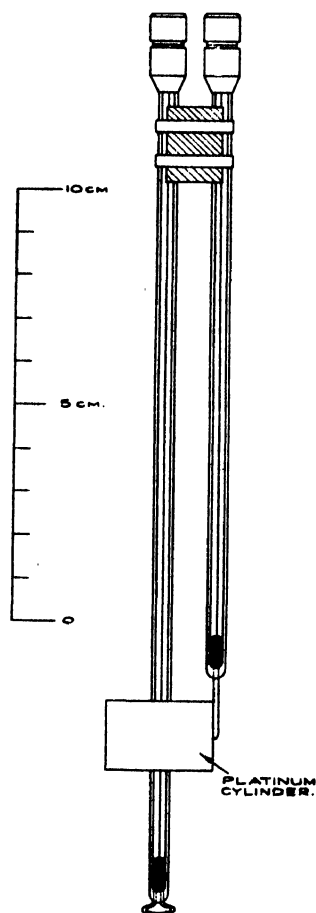


Fig. 1

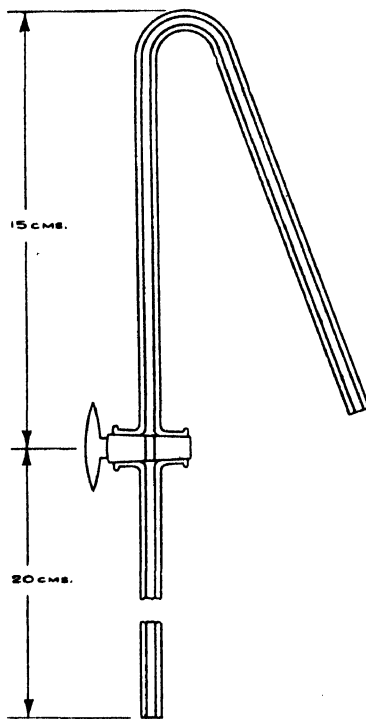


Fig. 2

The anode consisted of a cylinder made from platinum sheet, about 0.1 mm. thick, welded to a stout platinum wire lead. The cylinder, whose total surface area was approximately 30 sq.cm., was arranged to surround the lead to the cathode. The exact design of the anode is not important, and use could no doubt be made of other platinum ware which might be available. Carbon was tried in place of platinum for the anode, but the rods quickly disintegrated.

The only other special part of the apparatus was a siphon tube (Fig. 2), made of heavy-wall barometer tubing of 3 mm. bore and fitted with a capillary tap. The narrow bore of the tube, while not unduly restricting the flow, enabled a stable column of liquid to be maintained in the tube, even when the tap was closed, and the wide flat end of the tube made it possible for the liquid to be removed without any mercury being carried over.

The apparatus was arranged so that a number of electrolytic cells could be wired in series with a resistance to the 120-volt D.C. mains. There was a fall of 6-6.5 volts across each cell when a current of 8 amperes was flowing. No stirring arrangement was used, as the evolution of gas from the electrodes produced considerable agitation of the solution. Adjustment of the acidity of the solution during electrolysis was found to be unnecessary. At first the solutions were cooled during electrolysis, but it was found that if cooling was omitted the time for removal of 2 g. of nickel was reduced from 4 hours to about 1½ hours, and in the final procedure the time was further reduced to 1½ hours by heating the solution to boiling prior to the electrolysis. The heat developed in the process maintained a temperature of about 90° C. when a current of 8 amperes was used. Another result of the heating was that no trace of nickel could be detected in the electrolyte after 1½ hours, whereas complete removal of nickel was not achieved in 4 hours when cooling was employed.

A further reduction in the time of electrolysis from 1½ hours to ¾ hour may be obtained by reducing the volume of the electrolyte to 60 ml. The electrolyte is then contained in a 100-ml. beaker and the amount of sulphuric acid is suitably reduced. Details of the procedure finally adopted are as follows:

Experimental Procedure.—Two g. of the sample are dissolved in 15 ml. of nitric acid (1 : 1). The solution is cooled, 10 ml. of conc. sulphuric acid are added, the solution is evaporated until fumes of sulphuric anhydride appear, and is then heated for a further 10 minutes. The solution is allowed to cool, 50 ml. of water are added, and the liquid is boiled gently until all soluble sulphates dissolve. The hot solution is filtered into a 250-ml. beaker, the paper and any silica being washed with hot water. Cold freshly-prepared sodium hydroxide solution (20 per cent.) is added to the filtrate until a permanent turbidity is produced. The turbidity is cleared by the addition of dilute sulphuric acid, and the solution is finally acidified with 1 ml. of sulphuric acid (1 : 1) and made up to a volume of about 150 ml. It is then heated to boiling, 10 ml. of clean mercury are poured in, the electrodes and cover-glass are placed in position, and a direct current of 8 amperes is passed through the solution for 75 minutes. The electrolyte is then run off through the siphon tube into a 400-ml. beaker. The electrodes and the sides of the beaker are washed down with water, the washings being added to the main solution before the circuit is broken. Three washes of about 15 ml. each are sufficient to remove the last traces of electrolyte. Any mercury which may be carried over accidentally should be filtered off before proceeding.

Two g. of ammonium chloride are added to the solution, which is then heated nearly to boiling. A few drops of methyl red solution are added and ammonia solution is added carefully until the yellow colour of the indicator appears. The solution is then boiled for not more than 1 minute (*cf.* Blum, *J. Amer. Chem. Soc.*, 1916, 38, 1282), a little filter-paper pulp is stirred in, and the precipitate is collected on a Whatman No. 41 or equivalent paper. The precipitate is washed three times with hot ammonium nitrate solution (2 per cent.), and the paper and precipitate are returned to the beaker. Five ml. of hydrochloric acid are added, and the beaker is warmed on a water-bath for 5 minutes. One hundred ml. of hot water are added, and the aluminium is re-precipitated, the same procedure being used. When the aluminium-content of the sample exceeds 1 per cent., the second filtration may be carried out on a small Buchner funnel. The precipitate is washed 6 times with hot ammonium nitrate solution, dried and burnt off in a weighed platinum

crucible, with final ignition at 1200° C. for 30 minutes. The crucible is allowed to cool in a sulphuric acid desiccator, and the ignited precipitate is weighed rapidly as Al_2O_3 .

EXTENSION OF THE METHOD TO OTHER ALLOYS.—(i) *Nickel-aluminium "king" alloy.*—A "king" alloy containing approximately 50 per cent. of aluminium was analysed by the same process. The results are given in Table IV.

(ii) *Nickel-magnesium.*—The method has been applied to the determination of magnesium in nickel-magnesium alloys, nominally containing 0.3 per cent. of magnesium. The alloy (3.0 g.) is dissolved in 20 ml. of nitric acid (1 : 1), the solution is evaporated to fuming with 10 ml. of sulphuric acid, and the same procedure is followed as that used in the removal of nickel from the nickel-aluminium alloys. The magnesium is then precipitated as magnesium ammonium phosphate, ignited, and weighed as $\text{Mg}_2\text{P}_2\text{O}_7$.

Synthetic solutions containing 2 g. of nickel with 0.29 per cent. of added magnesium were analysed by this procedure. In three determinations the results were 0.30, 0.28 and 0.29 per cent. of magnesium respectively. In the last two instances the magnesium ammonium phosphate was re-precipitated, but the results suggest that normally this should not be necessary.

A "king" alloy containing about 18 per cent. of magnesium was analysed by the same method. Some results are given in Table IV.

(iii) *Nickel-beryllium alloy.*—A nickel beryllium alloy containing approximately 0.5 per cent. of beryllium was analysed by the electrolytic method.

1.5 g. of the alloy were dissolved in nitric acid, and after evaporation with sulphuric acid the nickel was removed by electrolysis. The beryllium in the electrolyte was then precipitated by the addition of ammonia solution in slight excess, litmus being used as the indicator. The precipitate was filtered, washed with ammonium nitrate solution, ignited and weighed as BeO . The results of duplicate determinations are given in Table IV.

TABLE IV

| Weight taken g. | Found | | Time of electrolysis, minutes |
|--------------------|-----------------------------------|-------------------|----------------------------------|
| | g. | Per Cent. | |
| Ni-Al | Al_2O_3 | Al | |
| 0.3 | 0.3046 | 53.7 _s | 30 |
| 0.3 | 0.3053 | 53.8 _s | 30 |
| Ni-Mg | $\text{Mg}_2\text{P}_2\text{O}_7$ | Mg | |
| 3.0 | 0.0401 | 0.30 (0.29*) | 100 |
| 0.5 | 0.4241 | 18.52 (18.45*) | 25 |
| Ni-Be | BeO | Be | |
| 1.50 | 0.0244 | 0.59 | 60 |
| 1.51 | 0.0243 | 0.58 | 60 |

* By cyanide method.

Effect of Manganese.—The nickel alloys under consideration contained no manganese, but, as the literature suggested that this element might be incompletely separated, an experiment was made to investigate this point. Manganese sulphate, in amount equivalent to 0.1 per cent. of manganese, was added to 2 g. of a nickel-aluminium alloy, the aluminium-content of which had been previously found to be 0.42 per cent. The solution was electrolysed, and the aluminium was determined in the usual way. The result was again 0.42 per cent., and the ignited precipitate of Al_2O_3 was perfectly white. The nickel-mercury amalgam was shaken for a few minutes with 30 ml. of nitric acid (sp.gr. 1.135), and the dissolved manganese was determined by the bismuthate method. Sixty-five per cent. of the added manganese was recovered, and, as only a small amount of the nickel dissolved in this treatment, it is presumed that the remainder of the manganese would have been recovered by further extraction of the mercury.

No manganese was noticed on the anode, and it would seem that no interference is to be expected from quantities of this order.

SUMMARY.—An investigation has been made into methods for the determination of aluminium in nickel-aluminium alloys.

The following methods have been compared:

Precipitation with:—(i) Ammonia in presence of potassium cyanide; (ii) ammonia and ammonium chloride; (iii) barium carbonate; (iv) sodium succinate; (v) ammonia after the removal of the nickel by electrolysis over a mercury cathode.

The last method has been found to be most satisfactory, and has been applied to nickel-aluminium alloys containing from 0.3 to 50 per cent. of aluminium.

The method has been found suitable also for the determination of magnesium and beryllium in nickel-magnesium and nickel-beryllium alloys.

A simple form of apparatus for the electrolysis is described.

REFERENCES

1. R. C. Chirnside, *ANALYST*, 1934, **59**, 278.
2. W. Gibbs, *Amer. Chem. J.*, 1883, **13**, 571.
3. E. F. Smith, "Electro-Analysis," London, 1919.
4. J. R. Cain, *J. Ind. Eng. Chem.*, 1911, **3**, 476.
5. G. E. F. Lundell, J. I. Hoffman and H. A. Bright, *id.*, 1923, **15**, 1064.
6. A. T. Etheridge, *ANALYST*, 1928, **53**, 423.
7. D. H. Brophy, *J. Ind. Eng. Chem.*, 1924, **16**, 963.
8. A. T. Etheridge, *ANALYST*, 1929, **54**, 141.
9. B. S. Evans, *id.*, 1935, **60**, 389.
10. G. E. F. Lundell and J. I. Hoffman, "Outlines of Methods of Chemical Analysis," New York, 1938, p. 94.

RESEARCH LABORATORIES OF
THE GENERAL ELECTRIC COMPANY LIMITED,
WEMBLEY, ENGLAND

February, 1940

DISCUSSION

Mr. C. E. BARRS recalled a useful paper by Withey (cf. *ANALYST*, 1916, **41**, 181) on the determination of various impurities in aluminium. In that method, use was made of the fact that tartaric acid would prevent the precipitation of aluminium by ammonium sulphide, whilst zinc and nickel were precipitated quantitatively. An important point in the present paper was that the alumina was very strongly ignited; it was essential to do that to obtain a satisfactory final weight.

Mr. SAXBY asked if the method could be applied to alloys used in magnet steel. Had Mr. Chirnside any method of differentiating between metal and oxide?

Mr. PINDER asked if the authors had tried the colorimetric method for these alloys; also, had oxine titration been tried?

Dr. B. S. EVANS, in a written communication, suggested that the separation of manganese described by the authors might have been a "fluke," as there was a definite back reaction. Manganese was incompletely deposited in all forms of apparatus other than that described by him in *THE ANALYST*. In his apparatus the separation was complete because the circulating cathode mercury was continuously treated to remove the deposited manganese. Deposition on the anode was a question of anode density and could be avoided by having that high enough. He suspected that, in the authors' experiments, the excess manganese remained in the solution and was not carried down to any extent with the alumina; it was not, if oxidation could be avoided. Etheridge had found only traces of manganese in the alumina, and determined them colorimetrically. Was this done in the authors' experiments? Referring to the determination of magnesium, he thought that it was dangerous advice to eliminate re-precipitation of the magnesium ammonium phosphate. He would refer the authors to Epperson's work, described in the *J. Amer. Chem. Soc.* in 1928 (*Abst. ANALYST*, 1928, **53**, 239).

Mr. CHIRNSIDE, replying, said that the ignition of aluminium oxide, to which Mr. Barrs had referred, was an important point. Above 1000° C. the conversion of γ to α alumina took place, and at 1200° C. was complete in a comparatively short time. The change to the α form not only ensured the complete expulsion of moisture but also resulted in a non-hygroscopic form of oxide for the final weighing. Mr. Saxby had mentioned magnet steels. The use of alloys containing aluminium for this purpose was growing, and he thought the method might be applicable to these alloys. The differentiation between aluminium present as metal and as oxide was an important point, and an investigation was being made on alloys to which known amounts of alumina had been

added. In reply to Mr. Pinder, the quantities of aluminium under consideration were considered to be above the normal range of the colorimetric determinations. Separations of nickel and aluminium by means of oxine had been tried but they had met with little success. In the case of nickel-magnesium alloys, having separated the nickel, he believed it would be satisfactory to determine the magnesium by means of oxine.

Written reply to Dr. Evans.—Dr. Evans very kindly read through the MS. of this paper and questioned whether the apparatus separated manganese. The nickel alloys did not contain manganese but the work described in the section on manganese was subsequently carried out for its general interest. All that was claimed as a result of these experiments was that no interference was found from manganese of the order of 0.1 per cent. The authors were well aware, from an extensive experience of silicate analysis, that manganese in small amount was not co-precipitated with aluminium hydroxide; the alumina precipitate in the experiment described was not tested for manganese. No manganese was deposited on the anode; some of it might have remained in solution, but it was possible that the conditions obtaining in the apparatus, though not required to effect the separation of manganese, might yet have done so.

They had tried to determine aluminium in nichrome some years ago, using Dr. Evans's apparatus. In his paper (*ANALYST*, 1930, **60**, 393) Dr. Evans admitted that he had not satisfactorily solved the problem of the composition of purifying solutions for the mercury (except in the case of manganese) and their failure to do so led to the abandonment of his method. The composition of the solution for extracting manganese was not given.

They were aware of Miss Epperson's work, and it was their usual practice to make two precipitations. However, they considered that the accuracy obtainable with one precipitation justified the suggestion made. Since the paper was written a number of gravimetric determinations involving the use of oxine had been made. In general, the results were a little higher than those obtained by the use of phosphate. This was a further reply to Mr. Saxby's question concerning the use of oxine.

Antimony in Tin-Antimony Oxides

By A. G. DUNBAR-POOLE

IN the investigation of the problem of the gravimetric determination of tin in brasses and bronzes,¹ it was found that when the procedure described was followed the final product consisted of the mixed oxides of tin and antimony present in the alloy.

The presence of antimony and, to some extent, its proportion in this mixture is indicated by the colour of the ignited mixed oxides. A precipitate in which the antimony-content is as little as one twenty-fifth of the tin present will give a definite pale blue colour. The intensity of this colour increases with rise in the proportion of antimony to tin, until when the tin/antimony ratio is about 1 : 10 the ignited mixture is bluish-black.

The ignited precipitate of tin and antimony oxides is practically insoluble in all single acids or mixtures of acids, and to determine the amount of antimony present it was found necessary to fuse the precipitate with caustic soda or Rose's mixture, to extract the melt and to precipitate the antimony as sodium antimonate, or to reduce it, in acidified solution, to the tervalent condition and determine it volumetrically.

Further experiments with this ignited precipitate have shown that it is possible to dissolve it by heating it in conc. sulphuric acid with addition of sodium thiosulphate. The antimony in the resulting solution will be in the tervalent condition and may be determined volumetrically by re-oxidation to the quinquevalent state—according to the method of Low.² The details of the procedure are as follows:

METHOD.—The ignited precipitate is transferred to a 400-ml. tall beaker, about 5 g. of sodium thiosulphate are added, and the mixture is heated on a hot plate until all the water of crystallisation has been expelled from the thiosulphate. During this process, when the salt is liquid, the beaker is rotated, so that intimate admixture of the oxides with the thiosulphate is effected. The beaker is then removed from the hot plate, 15 ml. of sulphuric acid (sp.gr. 1.84) are added, and

the beaker is replaced on the plate and heated until solution of oxides is complete. Heating for 20 to 30 minutes is generally sufficient to effect decomposition. The beaker is then removed and cooled, 100 ml. of cold water and 15 ml. of hydrochloric acid (sp.gr. 1.18) are added, and the whole is boiled for five minutes, after which 100 ml. of cold water are added and the solution is cooled by immersing the beaker in running tap-water.

When quite cold, the solution is titrated with $N/20$ potassium permanganate solution to a faint permanent tinge of pink (1 ml. $N/20$ $\text{KMnO}_4 \equiv 0.00305$ g. of Sb.).

COMPOSITION OF TIN-ANTIMONY OXIDES.—Experiments were made to ascertain the nature of these ignited tin-antimony oxides. An alloy of tin and antimony was made by melting together, under charcoal, "Chempur" tin and pure antimony. The ratio of tin to antimony was roughly 10 : 1. The antimony-content of the finely-rasped alloy was found by repeated direct volumetric determination, by the method of Low², and by analysis of the sulphides obtained from a solution of the alloy in *aqua regia*. The results ranged from 9.10 to 9.18 per cent.; mean, 9.15 per cent.

A one-gram portion of the alloy was digested in boiling nitric acid (sp.gr. 1.42), water was added, and the precipitate was allowed to settle, collected on an "ashless" filter-paper, washed with dilute nitric acid, and ignited in an electrically-heated muffle furnace. No trace of tin or antimony could be detected in the filtrate.

The precipitate was ignited in successive stages of two hours each by advancing the crucible to hotter portions of the muffle, free access of air being allowed. The weights recorded were as follows:—After ignition at 400°C ., 1.2850 g.; at 510°C ., 1.2832 g.; at 700°C ., 1.2815 g.; at 750°C ., 1.2810 g.; at 800°C ., 1.2810 g.

A further portion of the alloy was dissolved in *aqua regia*, the solution was rendered ammoniacal and then made acid with hydrochloric acid, and the sulphides of tin and antimony were precipitated with hydrogen sulphide gas. The sulphides were filtered off, washed, and ignited to oxides, which were finally roasted at about 750°C . The weight of oxides obtained was 1.280 g. per g. of the alloy, which is equivalent to 1.153 g. of SnO_2 , calculated on the basis of 90.85 per cent. in the alloy. This amount of tin oxide, deducted from the weight of ignited mixed oxides obtained, *viz.* 1.281 g. per g. of the alloy, leaves 0.128 g. for the antimony oxide compound. As the amount of antimony present (see above) was 0.0915 g. in 1 g. of the alloy, a factor of 1.40 is required to convert this amount into 0.128 g. of antimony-oxide compound.

This factor is found to agree with that required to convert Sb_2 into $\text{Sb}_2\text{O}_5 \cdot \text{H}_2\text{O}$. (If the most recently published atomic weights are used, the factor is 1.4023.)

Conversely, if the antimony exists in the ignited mixed oxides as $\text{Sb}_2\text{O}_5 \cdot \text{H}_2\text{O}$, the factor for conversion to Sb_2 being 0.7131, a figure of 9.13 per cent. of antimony is arrived at, which is in agreement with the amount (9.15 per cent.) found.

If the antimony in the ignited oxides was present as Sb_2O_3 , the factor for conversion into Sb_2 would be 0.7527 and would give a figure of 9.64 per cent. for the antimony content.

It is, seemingly, the opinion of many workers and authors that the antimony exists as Sb_2O_3 . In a very recent publication (W. W. Scott's *Standard Methods of Chemical Analysis*, 1939), the A.S.T.M. Methods of Chemical Analysis of Metals are given, and in Vol. II, p. 1357, Note 4, it is stated that: "In alloys containing an appreciable amount of antimony and in the presence of a large excess of tin, the antimony as determined by the method given under 'Analysis of Bronze Bearing Metal' should be calculated to Sb_2O_3 and a corresponding amount deducted from the weight of the ignited SnO_2 before calculating to tin."

In the present instance, if the antimony existed in the ignited mixed oxides as Sb_2O_3 (the factor for converting Sb_2O_3 to Sb_2 being 0.7919) the figure for antimony would be 10.14 per cent., or 1 per cent. more than is known to be present.

Still further proof was sought by analysing 0.5 g. of a gun-metal alloy, the

tin-content of which was 10.25 per cent. and in which the absence of antimony had been proved. To this 0.0110 g. of pure antimony metal was added, both were dissolved in *aqua regia*, and the exact procedure given for the determination of tin in bronzes and brasses¹ was followed.

The final precipitate of tin and antimony was ignited at about 800° C., and when constant weighed 0.0805 g. Conversion of the antimony present into $\text{Sb}_2\text{O}_3 \cdot \text{H}_2\text{O}$ gives 0.01545 g. and leaves 0.06505 g. for SnO_2 , which corresponds with 10.24 per cent. of tin.

The fact that these ignited mixed oxides are coloured tends to show that a definite compound is formed. Tin, in the form of metastannic acid or stannic sulphide, gives on ignition a white or grey-white oxide, SnO_2 . Antimony sulphide can be oxidised with nitric acid and ignited to give white Sb_2O_3 .

I prepared pure SnO_2 and pure Sb_2O_3 , ground these white oxides together in a mortar and re-ignited the mixture at about 800° C. for several hours, but no change in colour or weight was observed at the end of this period.

The conclusions are that when antimony is precipitated with metastannic acid and the precipitate is ignited, or, when the mixed sulphides of tin and antimony are ignited, the tin being in large excess, it would appear that the antimony exists in the form of a compound having the empirical formula $\text{SnO}_2 \cdot \text{Sb}_2\text{O}_3 \cdot \text{H}_2\text{O}$; that is, an antimoniate of tin which retains 1 mol. of water at temperatures between 700° and 800° C.

Even at temperatures higher than 800° C. there seems to be no further loss in weight of the compound. It was not possible, however, to attain in the electrically-heated muffle furnace used in these experiments a temperature higher than about 800° C. Stress must be laid on the desirability of having free access of air during ignition of this compound, especially when a gas-heated muffle is used.

I wish to thank the Director of Scientific Research of the Admiralty for permission to publish this paper.

REFERENCES

1. A. G. Dunbar-Poole, *ANALYST*, 1939, **64**, 870.
2. A. H. Low, *J. Amer. Chem. Soc.*, 1907, **29**, 66; *Abst.*, *ANALYST*, 1907, **32**, 101.
January, 1940

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

SUCCINIC ACID IN BEER

ON examining a range of beers from a number of breweries, by alkalisng 250 ml., evaporating to about 50 ml., acidifying, extracting with 250 ml. of ether and evaporating the extract, crystals appeared in three of these residues, which it was thought might be an unusual preservative. Larger quantities of the samples were extracted continuously with ether for 10 hours. The brownish extract contained masses of dark brown needles in small rosettes or clusters, the crystal form being quite distinct from those characteristic of salicylic or benzoic acid. The crystals were purified by recrystallisation from ether and washing with chloroform. They were then acidic, very soluble in water, less soluble in ether, and insoluble in chloroform, and melted at 183° C., which melting-point was not depressed on admixture with pure succinic acid. It was therefore concluded that the substance was succinic acid.

All the samples from which visible crystals separated were examined and found to contain succinic acid in amounts of the order of 0.002 to 0.008 per cent. w/v. It is to be expected that all beer contains succinic acid, but only occasionally is the amount sufficient to give visible crystals in an ethereal extract.

DOMINION LABORATORY
DEPT. OF SCIENTIFIC AND INDUSTRIAL RESEARCH
WELLINGTON, NEW ZEALAND

R. L. ANDREW
L. G. NEUBAUER

March 21st, 1940

A NEW METHOD FOR THE DETERMINATION OF FREE ACID IN SULPHATE OF ALUMINA, ALUM CAKE, ETC.

I HAVE pointed out (*Paper-maker*, 1940, 99, 14; *Special Export Number*) defects of existing methods for this determination and have shown that a rapid method of sufficient accuracy for industrial purposes can be based on the detection of high acidity in an aqueous solution of the sample by means of an indicator solution and subsequent titration with standard alkali back to the "neutral" point, provided that a suitable indicator be found.

Before such a method can be evolved it is necessary *inter alia* to establish a standard of "neutrality." Owing to crystallisation difficulties, it appears to be extremely difficult to obtain supplies of sulphate of alumina of the exact composition $\text{Al}_2(\text{SO}_4)_3 \cdot 18\text{H}_2\text{O}$. On the other hand, pure potash alum of the definite composition $\text{K}_2\text{SO}_4 \cdot \text{Al}_2(\text{SO}_4)_3 \cdot 24\text{H}_2\text{O}$ is easily obtainable, and I have shown that a solution of that salt, having the same $\text{Al}_2(\text{SO}_4)_3$ content as the required aluminium sulphate solution will have the same pH value. A solution of potash alum of equal $\text{Al}_2(\text{SO}_4)_3$ content is therefore used as a standard of "neutrality."

The most convenient strengths for the solution of the sample and the standard alkali are selected so as to avoid dilution sufficient in itself to cause pH changes or, on the other hand, concentrations high enough to cause temporary local precipitation of alumina.

The most convenient strength for the solution of the sample requires a "neutral" point of pH 3.5 to be covered by the indicator. As no suitable single indicator is available, a mixture of methyl orange and Martius yellow is used to cover the end-point. The indicator solution is prepared by grinding together 0.025 g. of methyl orange and 0.050 g. of Martius yellow with 50 per cent. industrial spirit until solution is complete and diluting to 100 ml. with the same solvent.

In the method finally adopted, 1 g. of the sample is dissolved in 100 ml. of water and the solution is titrated with *N*/4 sodium hydroxide solution, 4 to 6 drops of the indicator solution being added. A solution of 1.425 g. of potash alum in 100 ml. of water, with exactly the same amount of indicator added, is used as a blank. Towards the end of the titration, up to 30 seconds is allowed for the colour-change to develop. The number of ml. of the alkali solution required by 1 g. of the sample gives the percentage of free acid as SO_3 .

AYLESFORD PAPER MILLS
LARKFIELD, KENT

J. R. SIMMONS

June, 1940

Official Appointments

Erratum.—In the list of appointments on p. 411 (July issue) for "Westmorland County Council, Kendal Borough, W. H. Roberts" read "W. H. Roberts as a Public Analyst for that part of the County of Westmorland within the boundaries of the Borough of Kendal."
(C. J. H. Stock is the Public Analyst for the County of Westmorland.)

Ministry of Agriculture and Fisheries

CREAM CHEESES*

THIS Leaflet deals with the preparation and packing of cream cheeses. It is pointed out that the absence of legal standards of quality has resulted in a great variety of products, with a wide range of fat, being sold under the name of cream cheese.

The two main types sold in this country are:

DOUBLE CREAM CHEESE, which is made from cream containing about 50 per cent. of butter-fat, and

SINGLE CREAM CHEESE, which is usually made from cream containing 25 to 30 per cent. of butter-fat, and is thickened by adding rennet prior to drainage.

Methods of preparing these two types of cheese are described in detail.

Attention is directed to recent work (*J. Min. Agric.*, Sept., 1939), which has shown that cheese wrapped in close-fitting material (*e.g.* aluminium or coated tinfoil), keeps better than cheese packed in muslin or parchment paper; these promote the growth of mould.

* Advisory Leaflet No. 222, 1940. H.M. Stationery Office, York House, Kingsway, London W.C.2. Price 1d. net per copy, or 9d. per dozen copies.

General Medical Council

SECOND ADDENDUM TO THE BRITISH PHARMACOPOEIA, 1932*

THE Second Addendum to the British Pharmacopoeia, 1932, was published on June 14th, 1940, and became official from that date. The following new monographs are included:—*Emulsio Olei Morrhuae*, *Emulsio Olei Vitaminati*, *Extractum Malti cum Oleo Vitaminato*, *Liquor Vitamini A Concentratus*, *Liquor Vitamini D Concentratus*, *Liquor Vitaminorum A et D Concentratus*, *Oleum Amygdalae Volatile Purificatum*, *Oleum Hippoglossi*, *Oleum Vitaminatum*, *Toxinum Tetanicum Detoxicatum*. Amended formulae are given for *Linimentum Camphorae*, *Unguentum Acidi Tannici*, *Unguentum Aquosum*, *Unguentum Capsici*, and *Unguentum Hydrargyri Compositum*.

There are five appendices:—In IV.G the method of determining the ultra-violet absorption of cod-liver and halibut-liver oil at $328m\mu$, and of calciferol at $265m\mu$, is described.

Appendix X.C gives methods for determining iodine values (a) by the iodine monochloride method, (b) by the pyridine bromide method. It also describes the determination of the iodine value of the glycerides of halibut-liver oil.

In X.D. the S.P.A. method for the determination of unsaponifiable matter in fixed oils and fats is given.

In XI.C the determination of aldehydes in purified Volatile Oil of Bitter Almonds is described.

Appendix XV.O gives details for the assay of vitamin A by the spectrophotometric method at $328m\mu$. The factor declared by the Permanent Commission on Biological Standardisation of the League of Nations (at present accepted as 1600) is to be used for calculating the vitamin A content in Units per gram from the ultra-violet absorption figure.

Monographs of the British Pharmacopoeia, 1932, or of the Addendum, 1936, which are amended by this Addendum supersede, in their amended forms, the original monographs.

THE BRITISH PHARMACOPOEIA, 1932

ALTERATIONS AND AMENDMENTS

The Registrar of the General Council has sent us a notice to the effect that, in pursuance of Section 54 of the Medical Act, 1858, the General Council of Medical Education and Registration of the United Kingdom have caused to be altered and amended the British Pharmacopoeia, 1932, by modifying the list of medicines and compounds, and certain directions for preparing certain medicine and compounds, therein contained, as set forth in the following Schedule:

THE SCHEDULE

IPECACUANHA.—*Synonyms.*—*Ipecacuanhae Radix*: Ipecacuanha Root. British Pharmacopoeia, 1932, p. 237, line 4, "two-thirds" is changed to "three-fifths," so that the standard for alkaloidal content is amended to read:—"Not less than 2 per cent. of the total alkaloids of Ipecacuanha, calculated as emetine, of which not less than three-fifths consists of non-phenolic alkaloids, calculated as emetine."

EMPLASTRUM PLUMBI, Plaster of Lead.—*Synonyms.*—Lead Plaster; Diachylon Plaster; Diachylon.

INJECTIO HYDRARGYRI. Injection of Mercury. *Synonym.*—Mercurial Cream.

INJECTIO HYDRARGYRI SUBCHLORIDI. Injection of Mercurous Chloride. *Synonym.*—Calomel Injection.

UNGUENTUM HYDRARGYRI NITRATIS FORTE. Strong Ointment of Mercuric Nitrate. *Synonyms.*—*Unguentum Hydrargyri Nitratis*; Mercuric Nitrate Ointment.

Arachis Oil may be used, in place of Olive Oil, in making these four preparations.

* Pp. 22. Published by Constable & Co., Ltd., for the General Medical Council. 1940. Price 2s.

Medical Research Council

THE CHEMICAL COMPOSITION OF FOODS*

THIS Report supplements, but does not supersede, the three Reports (Nos. 135, 187 and 213) previously issued by the Council. The earlier Reports contained sections dealing with analytical methods, the losses caused by cooking, and the value of hemicelluloses and celluloses in animal nutrition, and reference may be made to them for information on these points. The new feature of the present Report is its classification of recent analytical data obtained by a systematised procedure.

The foods have been divided into arbitrary groups under the following headings: cereals and cereal foods; dairy products; meat, poultry and game; fish; fruit; nuts; vegetables; sugars, preserves and sweetmeats; beverages; beers; condiments; vegetable fats; cakes and pastries; puddings; cooked dishes; sauces and soups. The figures for the cooked dishes were obtained by computation, not direct analysis. The cereal group includes starchy foods such as arrowroot and tapioca, and Bovril, Oxo, Marmite and Virol have been grouped with the beverages. The tables are given in two series, the first showing the composition of the foods per 100 grammes and the second the composition per ounce. Edible material only has been analysed, but for foods such as fish and stone fruits, which are usually served with waste, the amounts of the edible constituents that would have been obtained from 100 g. and from 1 oz. of the food as served are also given. Vitamins do not come within the scope of the report.

As in former Reports, protein nitrogen has been differentiated from non-protein nitrogen in meat and fish and in mushrooms. Since the nitrogen/sulphur ratio in meat and fish has been found to be very constant (Master and McCance, *Biochem. J.*, 1939, 33, 1304), the sulphur in most of these foods has been calculated from the nitrogen figure.

The "available" phosphorus and "available" iron have been tabulated for a number of foodstuffs. By "available" phosphorus is meant the phosphorus not present as phytic acid (McCance and Widdowson, *Biochem. J.*, 1935, 29, 2694), and "available" iron has been taken as the iron which reacts with $\alpha\alpha'$ -dipyridyl (Shackleton and McCance, *Biochem. J.*, 1936, 30, 582; McCance, *Chem. and Ind.*, 1939, 58, 528; Abst., *ANALYST*, 1939, 64, 335).

NOTES ON TECHNIQUE.—For the determination of fat in malted foods the Soxhlet method gives low results; also, with many cereals it gives results lower than those obtained by von Liebermann and Szekely's method (*Pflüg. Arch. ges. Physiol.*, 1898, 72, 360). On the other hand, with condiments higher results are obtained by the Soxhlet method.

INDIVIDUAL FINDINGS.—Bovril contained more potassium (3.59 per cent.) than any other food examined. Parmesan cheese contained the largest proportion of calcium (1.22 per cent.), Marmite was richest in phosphorus (1.89 per cent.) and carrageen moss in magnesium (0.63 per cent.) and sulphur (5.46 per cent.). Liver contained the most copper (5.8 mg. per 100 g.), and curry powder the most iron (75 mg. per 100 g.). Of all the foods analysed, Gruyère cheese was richest in nitrogen (5.9 per cent.).

Points not discussed in the previous reports include the following:

- (1) The amounts of sodium and chlorine in packet cheese differ from those usually present in cheese.
- (2) Fried fish tends to contain more calcium than the fresh fish, owing to some of the small bones being included in the edible material of the former.
- (3) Glacé cherries contain much more iron than the raw fruit, probably owing to contamination during stoning.
- (4) Golden syrup contains 7 times as much sodium as chlorine; in black treacle the ratio is reversed.

CHEMICAL COMPOSITION OF COOKED DISHES WITH THEIR RECIPES.—A section of 13 pages, by C. M. Verdon-Roe, gives a series of standard recipes for cooked dishes containing several ingredients. These comprise preserves and sweetmeats, beverages, cakes and pastry, meat and fish dishes, egg and cheese dishes, sauces and soups.

Since pooled samples of all the ingredients of these dishes had been analysed, it was only necessary to determine the loss or gain of water during the cooking process to enable an average composition for the made dish to be calculated. Details of the procedure are given.

BIBLIOGRAPHY AND INDEX.—The Report concludes with a list of 22 references to works cited in the text and an index of 7 pages, giving the places in the tables of every food mentioned.

* Special Report Series, No. 235. By R. A. McCance and E. M. Widdowson. Pp. 150. H.M. Stationery Office, York House, Kingsway, London, W.C.2. 1940. Price 4s. net.

Eire

REPORT OF THE STATE CHEMIST FOR THE YEAR ENDED MARCH 31st, 1940

IN his Report Dr. T. S. Wheeler gives an outline of the chemical work carried out in the State Laboratory for all Departments of the Government. The total number of samples examined was 37,843, as compared with 40,240 in the preceding year. The main decrease was in the number of Revenue samples. On the other hand, there were substantial increases in the numbers of samples examined for the Departments of (a) Agriculture and Fisheries, and (b) Local Government and Public Health.

DAIRY VOLUMETRIC GLASSWARE.—Under Part IV, Sec. 32, Sub-sec. (1) (c) of the Dairy Produce Act, 1924, it is provided that dairy volumetric glassware is to be tested and stamped at the State Laboratory or, alternatively, at the National Physical Laboratory, Teddington. The total number of instruments tested was 5719.

SEA-WATERS.—In connection with the work of the International Council for the Exploration of the Sea, the Department of Agriculture and Fisheries carries out an annual investigation of sea-water around the Irish coast. Among the objects of the investigation is the determination of the relation of salinity to fish life. The salinities are determined by a standard method, which has been adopted by all the countries taking part in the investigation. The number of samples, taken at various stations, was 209, and the salinities lay, in general, between 34 and 35 parts of salt in 1000 of sea-water.

COMMODITIES FOR LOCAL AUTHORITIES.—The Minister for Local Government and Public Health is empowered under the Local Authorities (Combined Purchasing) Act, 1925, to have commodities for local authorities tested. The 213 samples submitted included tars, soaps, disinfectants, oils and paints; 12 did not comply with the requirements of the specification.

SALE OF FOOD AND DRUGS ACTS, 1875-1936.—Under Section 22 three samples were forwarded to the State Laboratory by direction of District Justices in cases in which the defendants had appealed against the certificates of local analysts.

MILK.—Under the Milk and Dairies (Special Designations) Regulations, 1938 and 1939, the Laboratory received 404 samples, 197 for chemical and 207 for bacteriological examination.

Federated Malay States

ANNUAL REPORT OF THE INSTITUTE OF MEDICAL RESEARCH FOR THE YEAR 1938

THE Report of the Director, Dr. A. Neave Kingsbury, gives an outline of the work of the different Divisions of the Institute, including those of Bacteriology, Chemistry, Entomology, Malaria Research, Pathology, Rat Virus Enquiry and the Serological and Medico-Legal Section.

The Chemical Division is under the direction of Mr. J. Shelton, F.I.C. Of the 5758 samples examined, 4442 were for the medical and health services and 524 for the police. The Health Officers submitted 1040 samples of milk, of which 14.3 per cent. were condemned as adulterated. These included 36 samples which complied with the legal standards, but were shown, by means of the Hortvet cryoscope, to contain added water.

LEAD IN CANNED FOOD.—The Sale of Food and Drugs Rules, 1938, prohibit the sale or importation of food in cans which are so constructed as to allow the contents to come in contact with any solder except the small amount necessary to seal a vent hole. Since these rules were introduced the quality of the containers has noticeably improved. Several samples, however, gave evidence that the cans had been used a second time.

LEAD IN SAMSU.—Twenty samples of samsu taken from the distilleries were examined, and were found to contain lead in amounts up to 3 p.p.m. As samsu is often stored in jars which quite probably have a lead glaze, it would seem advisable to examine retail samples as well as bulk samples from the distilleries.

SIZE OF SHOT IN CARTRIDGE.—In a case in which the police wished to know what size of shot had been fired from a shot gun cartridge, it was found that the topwad had retained an impression of the shot. From the marks it was possible to give an approximate idea of the size of the shot.

FORMIC ACID POISONING.—Five cases were investigated. In one of these, acid equivalent to 43 g. of anhydrous formic acid was isolated from the stomach and its contents. Formic acid of about 95 per cent. strength is used as a coagulant on rubber estates.

CAUSTIC SODA POISONING.—Of the 52 cases in which poison was detected, caustic soda was found in 18. Prior to 1934 very few cases of this type of poisoning occurred, but since then they have continued to increase. Steps are being taken to include caustic soda in the schedule of poisons.

IRRITANT POISON IN WELL WATER.—Vegetable matter taken from a well was identified as the fruit of a species of palm, probably *Caryota mitis* Lour. According to Gimlette ("Malay

Poisons and Charm Cures”) “The fruit of this palm is often put into wells with intent to cause annoyance. Bathing with well-water treated in this way gives rise to an intense itching of the skin and may cause an acute inflammation of the eyes.” This is said to be due to numerous fine needles of silica in the pericarp of the fruit.

TRANSFERENCE OF REVENUE STAMP.—One of the document cases investigated involved the question whether a revenue stamp had been removed from one document and placed upon another. Microscopical examination of the back of the stamp showed the presence of fibres of a certain type of paper, which were not found in the questioned document, but agreed with those of paper similar to that from which the stamp was thought to have been removed.

“GROUPING” OF BLOOD STAINS.—With a view to the further refinement of routine tests for the “grouping” of blood stains, preliminary investigations with anti-“M” and “N” sera have been made. Sera have been made and adsorbed. The results with experimental stains with 0.02 ml. of blood were inconclusive, but with double that quantity the reactions were more definite. During the year the police submitted about 600 exhibits of blood stains. The “A” and “B” grouping was attempted on 62 of these and conclusive results were obtained with 49.

RAT VIRUS ENQUIRY REPORT*

THE Rat Virus Enquiry was instituted at the request of the United Planting Association of Malaya to investigate the local possibilities of a commercial rat “virus” imported by air from Europe. Rats are responsible for much loss on coconut plantations and oil palm and rubber estates and also for disease among the workers.

All the local rats examined were “black” rats (*Rattus rattus*). No “brown” rats (*Rattus norvegicus*) were seen. In Kuala Lumpur the prevailing species is *Rattus rattus diardi*, while on the plantations the common species is *Rattus rattus jalorensis*. A few specimens of *Rattus concolor*, which is a very small rat, were also taken.

EXPERIMENTS WITH “VIRUSES.”—In order to ascertain if any of the known rat “viruses” could be effective against these local rats, “herd experiments” were undertaken. Rats were artificially inoculated with a particular organism and then introduced into a large “run” with a number of healthy rats. The artificial infection was effected in three ways: (a) intraperitoneal injection of the bacterial suspension; (b) injection into the stomach through a rubber tube; (c) feeding with milk in which the organism had been cultivated.

The organisms used in the experiments were:

- (1) Three strains of *S. enteritidis*:—*S. enteritidis* “Medan,” *S. enteritidis* “Liverpool” (from commercial “Liverpool Virus”) and *S. enteritidis* “Ratin” (from a commercial bacterial preparation).
- (2) Three organisms of the *Pasteurella* Group:—*P. muriseptica*, *P. cuniculiseptica*, *P. aviseptica*. These cause septicaemia in rats, if given in large doses, but are usually harmless to human beings.

From the results of the tests, which are described in detail, the conclusion was drawn that certain rat “viruses,” if applied in a suitable manner, were effective in killing local rats. With few exceptions, however, uninfected rats, after contact, under favourable conditions for the spread of infection, with infected rats, failed to become infected. It was therefore inferred that there was little possibility of the intentional precipitation of widespread epidemics among local rats by the use of these rat “viruses.”

EXPERIMENTS WITH ECTROMELIA VIRUS.—Ectromelia is a highly infectious disease of mice caused by a filtrable virus. This virus, obtained from the National Institute for Medical Research, Hampstead, proved pathogenic to local mice, but did not cause death, even when injected intraperitoneally in large amounts, into specimens of local *R. r. jalorensis*.

EXPERIMENTS WITH BACTERIAL TOXINS.—Sterile toxins of *B. dysenteriae*, *S. enteritidis* and *Cl. botulinum* were injected through stomach tubes into rats, but in no instance were pathological changes attributable to the respective toxins produced. The potency of the different toxins was checked by the inoculation of rabbits or guinea-pigs.

RATICIDAL VALUE OF COMMERCIAL VIRUSES AND CHEMICAL POISONS.—The results of inoculating or feeding rats with Liverpool Virus, New Liverpool Virus, Raxon, Ratinin, Vexterm Rat Biscuits, Ratbane and a number of chemical preparations are described in detail. Some of these poison baits proved effective. Bread soaked in Ratinin (which contains an extract of red squill) was readily eaten by rats when first offered, but was refused when subsequently supplied. A commercial phosphorus paste, containing 2 per cent. of phosphorus and 98 per cent. of inert ingredients (glucose, honey, flour) was readily eaten by rats, after it had been spread on cubes of bread, and proved effective. Ratbane, which consists of coarse, hard particles (possibly coarsely-ground corn), believed to be coated with zinc phosphide, proved very efficacious, although exposure diminished its attractiveness.

* Bull. No. 1. 1939. The Institute for Medical Research. By J. T. Paranjothy. Kuala Lumpur, 1939.

NON-COMMERCIAL CHEMICAL BAITS.—Four poison baits were prepared according to formulae recommended by the Department of Agriculture, Straits Settlements and Federated Malay States. They contained respectively 0.4 per cent. of thallium sulphate, 10 per cent. of sodium arsenite, 2 per cent. of arsenious oxide and 20 per cent. of barium carbonate.

A dose of 2 g. of any of the four baits may be considered a fatal one for local rats, since from 80 to 100 per cent. of the rats that ingested that amount died.

RELATIVE ATTRACTIVENESS OF BAITS.—Thirty rats were placed in a pen and supplied with 30 g. of each of the following baits: Vexterm biscuits (containing red squill), commercial phosphorus paste, thallium sulphate bait, sodium arsenite bait, arsenious oxide bait, barium carbonate bait. In one series of tests the rats were first starved for 24 hours, and in another series they were not starved but supplied with protein food in addition to the baits. After 24 hours, the baits were removed and weighed. Bread cubes with phosphorus paste proved the most attractive, and arsenious oxide and barium carbonate baits in the form of flour and bran balls were the least attractive. Thallium sulphate bait came second in both series of tests.

Various poison baits were exposed for 5 days to atmospheric conditions, but not to direct sun or rain. Bread smeared with pastes became rapidly unattractive, but loose hard substances such as Ratbane, lost attractiveness much more slowly. Rolled oats with thallium sulphate occupied an intermediate position.

THE ANTINEURITIC VALUE OF PARBOILED RICE*

RICE is the chief staple food in Malaya, and is consumed principally in three forms: (i) under-milled or husked rice; (ii) overmilled or polished rice; (iii) parboiled rice, made by soaking the padi in water, boiling it until the grains burst, and drying it in the sun. When properly prepared, parboiled rice is nearly translucent and varies in colour from almost white to light yellow.

Vedder and Feliciano (*Philip. J. Sci.*, 1928, 35, 351; Abst., ANALYST, 1928, 53, 542) classified milled rice roughly, according to the amount of pericarp left on the grain, into (a) highly milled rice with 0 to 20 per cent. of external layers present; (b) medium-milled rice, with 21 to 49 per cent.; (c) undermilled rice, with 50 to 100 per cent. The danger of beri beri, resulting from the consumption of milled rice, largely depends on the amount of pericarp layers removed. When undermilled rice is used, beri beri does not occur: and with medium-milled rice, if cases occur at all, they are apt to be sporadic.

Typical samples (158 in all) of the forms of rice consumed by the rural Malay population were examined by Vedder's iodine test (*loc. cit.*) to determine the approximate percentage of pericarp remaining on the grains. Of these samples, 23 (15 per cent.) had between 50 and 100 per cent. of pericarp remaining (undermilled), 81 (51 per cent.) had between 21 and 49 per cent. (medium-milled), and 54 (34 per cent.) had 20 per cent. or less (overmilled).

In addition to vitamin B₁, the major portion of the fat, proteins and mineral salts (especially calcium and phosphorus) is also concentrated in the pericarp layers. Thus Sreenivasan (*Ind. Med. Gaz.*, 1939, 74, 35) has shown that the outer envelope or bran and germ together contain from 50 to 75 per cent. of the mineral matter of the grain, more than 25 per cent. of the proteins, and practically the whole of the vitamins and fat.

It is now generally accepted that the antineuritic value of parboiled rice, although milled, is much greater than that of raw milled rice. It was suggested by Aykroyd (*J. Hyg.*, 1932, 32, 184) that during the process of parboiling some of the vitamin B₁ (and possibly also mineral matter and protein) is dissolved out of the pericarp and absorbed by the endosperm, thus making the vitamin B₁ content of milled parboiled rice considerably less dependent on the amount of pericarp retained. This hypothesis was supported by the fact that the polishings from parboiled rice were considerably poorer in vitamin B₁ and phosphate than the corresponding polishings from raw rice.

More recently, Subrahmanyam, Sreenivasan and Das Gupta (*Ind. J. Agric. Sci.*, 1938, 8, 459) have proved that this absorption of proteins and mineral salts by the endosperm at the expense of the integuments does, in fact, occur.

The fact that undermilled rice deteriorates rapidly on storage, developing an unpleasant odour and taste, has made it much less popular than the nutritionally inferior polished grain. Parboiled rice is in a different position. If properly prepared, it can be almost as white and palatable as polished rice. Unfortunately, however, the commercial article is frequently of bad colour and may have an objectionable flavour. These drawbacks are largely due to incorrect methods of preliminary soaking, which encourage the development of anaerobic bacteria. On the other hand, the steaming process is likely to sterilise the grain, and, provided that the final drying is sufficient, there is no reason why parboiled rice should not be capable of storage for long periods without serious deterioration.

The feeding experiments on rats, described in detail in the Report, show that the antineuritic values of parboiled rice, whether overmilled or undermilled, compare very favourably with those of undermilled raw rice.

* Bull. No. 4. 1939. The Institute for Medical Research. By I. A. Simpson. Kuala Lumpur. 1940.

Investigation of Atmospheric Pollution

REPRESENTATIVES of local authorities and other bodies co-operating in the investigation of atmospheric pollution met, by courtesy of the London County Council, in conference at the County Hall on May 28th. Among others, representatives of Cardiff, Dagenham, Glasgow, London County, Halifax, Manchester, Newcastle, Salford, Sheffield, Rotherham, Westminster, the British Commercial Gas Association and Cadbury Brothers, were present.

The Conference, in considering its annual report to the co-operating bodies, unanimously agreed that while contribution to the war effort was the first duty and desire of every organisation, the need for vigilant attention to the purity of the atmosphere had by no means decreased since the outbreak of the war. The wasteful burning of fuel and the detriment which pollution caused to the nation's health were stressed as two aspects of the problem which must not be forgotten, particularly in wartime. The Conference therefore urged all local authorities to do whatever lay in their power to maintain the Investigation.

At the close of the meeting, Professor W. H. Roberts, M.Sc., F.I.C., of Liverpool, was unanimously elected Chairman of the Conference, in succession to Alderman Adams, M.P., of Newcastle, who had held the office for three years in succession.

Royal Agricultural Society of England

ANNUAL REPORT OF THE CONSULTING CHEMIST FOR 1939

AMONG subjects of interest to the farming community, commented upon by Mr. Eric Voelcker in his Report, are the following:

SOYA-BEAN HUSKS.—A sample of soya-bean husks, offered at £5 17s. 6d. per ton, gave the following analytical results:—moisture, 11·51; oil, 4·10; albuminoids, 15·87; carbohydrates, etc., 38·30; woody fibre, 25·20; mineral matter (ash), 5·02 per cent. The sample contained 2·54 per cent. of nitrogen, and the mineral matter included 0·55 per cent. of sand and siliceous matter. This sample was inferior to others examined because of the low percentage of oil and high fibre-content, and the price appeared excessive. One sample gave 8 per cent. of oil, 23 per cent. of albuminoids and 17 per cent. of fibre. A good sample of soya-bean husks is not equal to pea meal, but is better than undecorticated cotton-seed meal, and would make a useful addition to a feeding ration; with such a high fibre-content the material is not suitable for pig-feeding.

"OAT FEED."—A sample submitted contained, besides oat husk, some barley husk together with tapioca meal. As its fibre-content was 22·02 per cent., a more apt name for it would have been oat husk refuse. In this connection reference is made to a case in which a firm was convicted and fined for adding not less than 10 per cent. of oat shud to ground oats. There is no statutory limitation to the amount of fibre in ground oats, and, although the Act defines Ground Oats as "the meal obtained by grinding commercially pure oats as grown," the defendant argued that, as only a product of the oat had been added, no offence had been committed. An average figure for the fibre-content of oats may be taken as about 10 per cent., and when one finds a figure considerably in excess of that proportion, it is reasonable to assume that an addition has been made. In this particular case the defendant admitted the addition of 2 cwt. of oat shud to each ton of pure oats. Such adulteration is not apparent to the eye, and farmers should be on their guard against it.

REFUSE FROM A BACON FACTORY.—A sample submitted had the following composition:—moisture and organic matter, 74·98; phosphoric acid, 5·36; lime, 6·79; salt, 3·50; magnesia, iron oxide, etc., 4·05; sand and siliceous matter, 5·32 per cent. The amount of nitrogen in the moist organic matter was equivalent to 8·11 per cent. of ammonia and the oil therein was 8·54 per cent. Although the oil-content was fairly high, it did not appear to be excessive for manuring purposes. Such a material would be a useful source of organic matter when dug into the soil.

COMPARATIVE ANALYSES OF RED AND WHITE CARROTS.—The following percentage figures were obtained:

| | Water | Oil (pet. spt. extract) | Albu- minoids | Sugar | Other sol. carbo- hydrates, etc. | Woody fibre | Mineral matter (ash) |
|--------------|-------|-------------------------------|------------------|-------|--|----------------|----------------------------|
| Red carrot | 87·79 | 0·16 | 1·26 | 3·00 | 5·88 | 0·81 | 1·10 |
| White carrot | 88·31 | 0·08 | 0·91 | 3·00 | 5·78 | 0·85 | 1·07 |

As the red carrot was little richer than the white, whilst the yield from the latter was three times that of the former, it was proposed by the member submitting the samples to grow only the white variety. Further examination of the two varieties, however, showed that the white carrots contained only 0·8 mg. of carotenoids per kilo. (not calculated on dry matter), whereas the red carrots contained 180 mg. per kilo. It would therefore seem inadvisable to replace the red by the white variety solely for the reason that the latter gives a much higher yield.

British Standards Institution

WAR EMERGENCY STANDARDS.—A communication from the Director points out that, owing to war conditions, the requirements of some existing British Standard Specifications cannot be strictly complied with by the manufacturers, who may in consequence be compelled to contract out of them. The British Standards Institution is meeting this situation by undertaking the revision of such Specifications and their issue as War Emergency Revisions, and the preparation of War Emergency Standards. These War Standards and Revisions are published on yellow paper to avoid confusion with the general B.S. standards, and it is made clear, wherever necessary, that they apply only to the home market and not to the export trade and can be modified again when peace comes. A number of such War Revisions have already been issued. They are put through with the minimum of delay—in some cases within a few days.

THE following British Standard Specification has been prepared by the Institution at the request of the Air Raid Precautions Department of the Ministry of Home Security.

BS/A.R.P. No. 40—1940. BLEACH OINTMENT (ANTI-GAS OINTMENT No. 1).*

The ointment shall consist of equal parts by weight of bleaching powder and white mineral jelly, and must be in the form of a uniform smooth paste, free from foreign matter and visible impurities. Undue heating must be avoided during mixing.

The bleaching powder used must be suitably stabilised, free from visible particles, and 99 per cent. must pass through a 60-mesh B.S. test sieve. It must contain not less than 30 per cent. of available chlorine and should not lose more than one-fortieth of its original chlorine-content after exposure for 2 hours at 100° C. in a specified manner.

The white mineral jelly shall comply with the following standards:—flash-point, not lower than 176·7° C.; drop-point, not lower than 40° C.; ash, not more than 0·03 per cent.; loss on heating for 6 hours at 100° C., not more than 1 per cent.; freedom from acidity, determined by boiling a mixture of 5 g. with 10 ml. of alcohol and testing with methyl orange. The jelly must also answer tests for freedom from reactive compounds (*e.g.* unsaturated hydrocarbons). Further tests include a check of the available chlorine in a freshly prepared sample of the ointment (not less than 14 per cent.); heating of the ointment for three hours at 57° C., when the temperature should not exceed 62° C., followed by a further check of the available chlorine, which should not be more than 2 per cent. below the original figure. Appendixes to the specification give the methods of determining (i) coarse particles in the ointment, (ii) available chlorine in the ointment, (iii) available chlorine in bleaching powder, (iv) stability of bleaching powder at 100° C., and (v) stability of the white mineral jelly.

The available chlorine in a freshly prepared sample of bleach ointment must not be less than 14 per cent. The labels and containers must be marked with the name "Bleach Ointment (Anti-gas Ointment No. 1)," the name of the maker, and the date of manufacture.

The following British Standard Specifications have also been published†:

No. 593—1940. GENERAL PURPOSE LABORATORY THERMOMETERS.

This is a revised Specification. It includes five series of thermometers for general purposes, the chief change in this revision being the inclusion of a series E, the thermometers in which are specially chosen to be suitable for use with distillation flasks specified in B.S. No. 571. The other series specified include thermometers for both Centigrade and Fahrenheit scales for partial and for complete immersion.

No. 611—1940. PETRI DISHES.

This Specification has recently been revised. The revision is mainly concerned with the tolerances, but the height and thickness of the top and bottom dishes have also been revised.

* Copies of the full specification can be obtained from the British Standards Institution, 28, Victoria Street, London, S.W.1. Price 2d. net.

† Obtainable from the Publications Department, 28, Victoria Street, London, S.W.1. Price 2s. net. Post free 2s. 2d. each.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Constitution of Banana Starch. E. G. E. Hawkins, J. K. N. Jones and G. T. Young. (*J. Chem. Soc.*, 1940, 390-394.)—Banana starch is best prepared by shaking minced, peeled unripe bananas in succession with alcohol (twice), and then with an aqueous 1 per cent. solution of sodium sulphite. If the fibrous matter is separated on a coarse muslin filter, the starch is deposited from the filtrate, and may be collected on a fine cloth filter or by flotation in alcohol. It forms a clear paste in hot water, which gives the deep-blue iodine reaction and does not reduce Fehling's solution. One preparation gave $[\alpha]_D^{20} + 152^\circ$ in *N* sodium hydroxide solution; P_2O_5 , 0.06; N, 0.15 per cent.; acidity equiv. to 0.6 ml. of 0.1 *N* alkali per g. Hydrolysis with taka-diastase left 5 per cent. of fibrous residue, and the reducing value of the residual solution corresponded with 98 per cent. of the theoretical amount of glucose. Distillation with 12 per cent. hydrochloric acid yielded no furfuraldehyde, and the amount of carbon dioxide evolved corresponded with the decomposition of hexose and, therefore, with the absence of uronic acids (*cf.* Campbell, Hirst and Young, *Nature*, 1938, 142, 912). After hydrolysis with 2 per cent. sulphuric acid the reducing power was 97 per cent. of the value corresponding with quantitative hydrolysis to glucose, an 88 per cent. yield of crystalline glucose being obtained from this solution. In general appearance, granule size and paste-forming properties, banana starch, as thus prepared, resembles potato starch. The preparation of the fully-substituted acetyl and methyl derivatives is more difficult than for potato starch, but the properties of the corresponding derivatives of the two starches are similar. Methylated banana starch (prepared directly, or by methylation of the acetate) was shown by viscosimetric methods (*cf.* Hirst and Young, *id.*, 1939, 1471, 1475) to have a mol. wt. of about 200,000. Hydrolysis of these methyl derivatives resulted in 2,3,4,6-tetramethyl glucose, 2,3,6-trimethyl glucose and dimethyl glucoses only, in amounts corresponding with the presence of a repeating unit of about 24 glucose residues. The molecular structure of banana starch closely resembles that of rice starch in respect of the "disaggregation reactions" of the methyl derivatives, *i.e.* the hydrolysis (*e.g.* by 1 per cent. oxalic acid at 60° and 75° C.) of the bonds between repeating units to give products of lower mol. wt. but unchanged chain-length. Throughout this operation the proportion of end-group remains unchanged, the solution is non-reducing and the rotation is undiminished, as distinct from the formation of reducing dextrans and, ultimately, of glucose, which

results from normal acid hydrolysis. Consideration of the kinetics of the disaggregation reaction shows that a normal glycosidic bond links the repeating units, so that under identical conditions the 1,6-fructofuranoside linkages in methyl inulin are hydrolysed about 7 times more rapidly than the bonds between the repeating units in methylated starches; this is illustrated graphically. Full experimental details of the methods of hydrolysis, methylation and disaggregation are given. J. G.

Determination of Amino-nitrogen in Malt Extracts. S. R. Snider. (*Cereal Chemistry*, 1940, 17, 121; *J. Inst. Brewing*, 1940, 46, 154-155.)—A method based on Pawlowski's modification of formal titration is as follows:—Sixty ml. of the wort or extract (12.5 per cent.) in a 100-ml. flask are kept in boiling water for 10 minutes, cooled, diluted to 100 ml. after addition of 10 ml. of 20 per cent. barium chloride solution and 5 ml. (7 ml. for proteolytic extracts) of saturated barium hydroxide solution, and filtered through a covered funnel after standing for 30 minutes. Four 20-ml. portions are measured into 125-ml. flasks, which are kept closed with rubber stoppers. Into flasks (1), (2), (3) and (4) are measured respectively 2 ml. of 0.02 per cent. phenol red solution, 2 ml. of water, and two 2-ml. portions of 0.04 per cent. thymol blue solution. The contents of flask (1) are then titrated with *N*/10 hydrochloric acid to a distinct, permanent yellow (pH 6.8). The same amount of acid is put into each of the remaining three flasks, followed by 10 ml. of 36-38 per cent. neutral formaldehyde (pH 5.6); cloudy or inferior grades should not be used. After titration of (3) with *N*/10 sodium hydroxide solution until it nearly matches a thymol blue pH 9 colour standard, and addition of an equal amount of the alkali to (2), the colours of (3) + water cell, and of (2) + thymol blue colour standard are compared in a colorimeter, a daylight lamp being preferable to daylight. If the matching is close, the titration of (3) is completed to an exact match after dilution of (2) and (3) to 40 ml. each. Flask (4) is used for a duplicate titration. Matching is more sensitive if colour standards are used on both sides of the titrated solution in the colorimeter; addition of 0.1 ml. of *N*/10 sodium hydroxide solution should make a matched solution perceptibly darker than the standard. A blank titration on the reagents is made, with water instead of wort or extract, while water cells replace the compensating wort solution in the colorimeter. If *T* and *B* represent the ml. required by the test solution and in the blank titration, and *M* the mg. of wort or extract in the 20 ml. titrated, the percentage of amino-nitrogen in the sample is $140 (T-B)/M$. E. B. D.

Keeping Properties of *Hydnocarpus Wightiana* Oil and its Derivatives. U. P. Basu and A. Mazumdar. (*J. Indian Chem. Soc.*, 1940, 17, 280.)—The inhibition of autoxidation of *Hydnocarpus Wightiana* oil, which can be brought about by addition of 7 per cent. of creosote (*Leprosy in India*, 1939, 11, 53) is shown still to occur if the concentration of the creosote is reduced to 0.1–0.2 per cent. The unsaponifiable matter of the oil, about 0.02 per cent., is not pro-oxygenic in nature. A concentrate prepared by a slight modification of the method of Green and Hilditch (*J. Soc. Chem. Ind.*, 1937, 56, 237; Abst., ANALYST, 1937, 62, 206), when dissolved in bicarbonate solution, reduced Fehling's solution, contained nitrogen and was insoluble in ether, but had no anti-oxygenic power. A sesame cake preparation, however, which was rich in nitrogen (6.1 per cent.), had a considerable anti-oxygenic effect. Thus the peroxide value (63.8 in terms of 0.01 N thiosulphate solution), obtained by heating an oil at 100° C. for 4½ hours, was reduced to about 2.6 when 0.5 per cent. of sesame concentrate was mixed with the oil prior to the heating. D. G. H.

Neem Oil I. M. Qudrat-i-Khuda, S. K. Ghosh and A. Mukherjee. (*J. Indian Chem. Soc.*, 1940, 17, 189–194.)—The oil was expressed from Birbhum neem seed (*Melia azadirachta*). Volatile substances were removed by prolonged steam-distillation of 30 lbs. of crude oil, the distillate consisting of the odoriferous constituent of the oil mixed with some solid material. On repeated distillation of the oil 15 g. of a colourless mobile oil with a penetrating, unpleasant odour were obtained; its probable molecular formula was $C_{18}H_{30}O_2S$, and it was termed "neemola." The residue of oil from the steam-distillation was extracted with several litres of water, and the bitter principle was isolated from the extract in the form of a solid substance termed "margosin," which, after purification, was found to be a glucoside with the formula $C_{30}H_{48}O_{10}$. The fatty oil remaining after removal of neemola and margosin was saponified and yielded a mixture of four acids which appeared to be different from those described by Roy and Dutt (*J. Soc. Chem. Ind.*, 1929, 48, 3337; Abst., ANALYST, 1930, 55, 50; and by Child and Ramanathan (*id.*, 1936, 55, 1247; Abst., ANALYST, 1936, 61, 498). One acid, m.p. 67° C., appeared to be an isomer of tetradecic acid, $C_{14}H_{28}O_2$, and was called neem acid A. Another acid, neem acid B, regarded as a higher homologue of acid A (m.p. 55° C.), is described as an isomer of palmitic acid. After separation of these two solid acids a mixture of a solid and liquid oil was obtained on distillation under reduced pressure of the solid acidic substance remaining in the mother liquor. The methyl esters were fractionated into two distinct portions; the lower-boiling ester yielded neem acid C, a solid unsaturated acid ($C_{18}H_{30}O_2$) of the oleic acid series, and the higher-boiling ester produced neem acid D ($C_{18}H_{32}O_2$), semi-solid at ordinary temperature

and apparently belonging to the cyclic acids. It is perhaps similar to *hydnocarpus* or *chaulmoogric* acid. D. G. H.

Determination of Indole by a Modification of Ehrlich's Reaction. L. H. Chernoff. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 273–274.)—Ehrlich's reaction (*Deut. Med. Woch.*, 1901, 1) depends upon the formation of a pink colour when indole is treated with dimethylaminobenzaldehyde in presence of hydrochloric or sulphuric acid. Separation by steam distillation of indole from products containing it is usually considered necessary, but the method to be described avoids steam distillation and appears to be conveniently applicable to food and biological material. It has been applied by Clarke *et al.* (*J. Assoc. Off. Agr. Chem.*, 1937, 20, 475; Abst., ANALYST, 1937, 62, 806) to butter and to distillates from butter. It is based primarily on the observation that when a solution of indole in chloroform is treated with dilute acid (up to about 12 per cent.) and Ehrlich's reagent the colour remains in the chloroform layer, but if the test is made with stronger HCl the colour is transferred to the aqueous phase. In the method described syrupy phosphoric acid is used instead of hydrochloric acid, because it is more easily separated from mixtures with chloroform and is less liable to promote interfering reactions with other substances. The reagent is prepared by dissolving 0.2 g. of purified dimethylaminobenzaldehyde in 100 ml. of 85 per cent. phosphoric acid and the test is made as follows: The reagent (5 ml.) is added to 50 ml. of chloroform containing from 1 to 10% of indole in a separating funnel and, after the mixture has been shaken for 2 minutes, 25 ml. of glacial acetic acid are added. After further shaking the mixture is allowed to stand. The lower layer is transferred to a Nessler glass, and the chloroform is washed with 2.5 ml. of phosphoric acid, which is added to the first extract. The liquid is then diluted to 50 ml. with glacial acetic acid, and the colour produced is directly proportional to the amount of indole present. Standard comparison solutions from indole treated in this manner are stable for 1 or 2 days. The comparison may be made in a Clifford neutral wedge photometer (U.S. Food and Drug Administration, Food Control Statement, 1935, 41, 1). The curve shows a maximum value in the neighbourhood of 570m μ , and as little as 0.5% of indole may be detected. The acetic acid used must be free from formaldehyde, and the chloroform should be washed free from alcohol. As indole is apparently polymerised by phosphoric acid, the dimethylaminobenzaldehyde reagent must be added first to the chloroform solution. Indole may be extracted with chloroform from slightly acid or slightly alkaline solutions. Biological material may be made slightly alkaline with dilute sodium hydroxide solution and extracted with a measured volume of chloroform. The separated chloroform layer is then washed with a little dilute hydrochloric acid, and an aliquot portion is treated in the

manner described. Emulsions formed during alkaline extractions may be broken by the addition of finely powdered ammonium sulphate, which also serves to remove formaldehyde.

A. O. J.

Mesul: The Bitter Principle of *Mesua ferrea*. P. Dutt, N. C. Deb and P. K. Bose. (*J. Indian Chem. Soc.*, 1940, 17, 277-279.)—Nageshwar oil, expressed from the seeds of *Mesua ferrea* Linn., N. O. *Guttiferae*, deposits "stearine," which has been found to contain a crystalline substance soluble in aqueous alkali solutions to form a deep yellow solution; this is responsible for the yellow colour observed by Chatterjee and Gupta (*Oil and Colour Trades J.*, 1937, 91, 1656), which is produced when the oil comes in contact with dilute alkali. Nearly 1 per cent. of pale yellow transparent prisms, m.p. 154° C., was isolated. This substance, called "mesul," is neutral to litmus, has a somewhat bitter taste, and is soluble in 1 per cent. aqueous caustic alkali solution, in 10 per cent. ammonia and in hot sodium carbonate solution, but is insoluble in sodium bicarbonate solution. It is precipitated unchanged from alkaline solution by acids, and in alkaline medium gives a dirty green colour with ferric chloride; it is phenolic in character. It is optically inactive, free from methoxy- and methylenedioxy-groups, and is not a hydroxy-flavone. The analytical and molecular weight data allow of two formulae: $C_{25}H_{22}O_6$ or $C_{25}H_{20}O_6$, but in view of the analytical data for dimethylmesul, the first is the more probable. Dimethylmesul contains a lactone ring, but gives no colour with ferric chloride.

D. G. H.

***Cannabis indica*. Isolation of Cannabidiol from Egyptian Hashish. Observations on the Structure of Cannabinol.** A. Jacob and A. R. Todd. (*J. Chem. Soc.*, 1940, 649-653.)—A fresh specimen of Egyptian hashish has been examined by the method used previously for Indian hashish (*Biochem. J.*, 1939, 33, 123; *Abst. ANALYST*, 1939, 64, 208), and cannabinol and cannabidiol (a typical constituent of American hemp resin) have been found to be present in approximately equal quantities. So far, cannabidiol has not been isolated from Indian resins, nor cannabinol from American resins. The experimental work supports the view of American authors (Adams and Clark, *J. Amer. Chem. Soc.*, 1940, 62, 196) that cannabidiol is a double unsaturated derivative of menthylbenzene containing two OH-groups and one *n*-amyl group at positions in the benzene ring corresponding with those substituted in cannabinol. Both cannabinol and cannabidiol appear to be inactive in the Gayer test on rabbits. (*Arch. exp. Path. Pharm.*, 1928, 129, 312.)

D. G. H.

Biochemical

Inertia of Highly Unsaturated Fatty Acids in the Animal, Investigated with Deuterium. K. Bernhardt and R. Schoenheimer. (*J. Biol. Chem.*, 1940, 133, 707-

712.)—The manner in which doubly and trebly unsaturated fatty acids are formed in the body was studied by feeding mice on a fat-low diet of bread and heavy water. The saturated and total unsaturated fatty acids isolated from the animals were found to contain considerable amounts of deuterium, confirming the results of earlier experiments; the saturated acids contained more deuterium than the unsaturated acids. The unsaturated acid fraction yielded α -linolic acid as the tetrabromide, and a trebly unsaturated acid as the hexabromide. None of these highly unsaturated acids contained more deuterium than could be accounted for by the error of the analysis. It is concluded that such acids, unlike the saturated and less unsaturated acids, are not biochemically synthesised, but are derived directly from the diet. The results indicate, furthermore, that hydrogen of CH_2 , CH_3 , and CH groups is not exchangeable with the hydrogen of the water of the body fluids.

F. A. R.

Biuret Reaction in the Estimation of Serum Proteins. I. A Study of the Conditions necessary for the Production of a Stable Colour which bears a Quantitative Relationship to the Protein Concentration. H. W. Robinson and C. G. Hogden. (*J. Biol. Chem.*, 1940, 133, lxxx-lxxxii.)—Absorption curves of the colour produced in the biuret reaction with serum proteins and varying concentrations of sodium hydroxide and copper sulphate were made, and the colour densities at wave-lengths of 560 and 700 $m\mu$ were compared with the nitrogen contents of the solution. A colour that remained constant for at least 48 hours was obtained by the following method:—Serum is precipitated with trichloroacetic acid and centrifuged; the precipitate is dissolved in 3 per cent. sodium hydroxide solution, and the colour is developed with copper sulphate solution. Under these conditions the colour densities are proportional to the amounts of protein present. A wide variation in the concentration of copper had no influence on the colour. Rabbit serum diluted with saline was a more satisfactory standard than dog or human serum, and showed no loss of protein after being stored in the refrigerator for 6 months. By filtering the coloured solution a reduction in the intensity was obtained, but the amount of colour was still proportional to the nitrogen-content; it seems probable that the estimation can be carried out in a photoelectric colorimeter without the use of a standard.

F. A. R.

Colorimetric Test for Methionine. M. X. Sullivan and T. E. McCarthy. (*J. Biol. Chem.*, 1940, 133, c-ci.)—Five ml. of a solution containing 1 mg. of methionine in water or in 0.1 N hydrochloric acid are treated with 0.3 to 1.0 ml. of a 4 per cent. aqueous solution of sodium nitroprusside, and the solution is made alkaline with sodium hydroxide solution. After 5 minutes the yellow solution is acidified, and then becomes a pronounced red. Of the

ordinary amino acids, only histidine gives a red colour, and methods of overcoming interference from this source are being explored.

F. A. R.

Effect of the Oral Administration of Dimethylaminoazobenzene (Butter Yellow) on the Growth of the Rat. J. White. (*J. Biol. Chem.*, 1940, 133, cviii.)—When 60 mg. of butter yellow per 100 g. of basal diet were added to the diet of young growing rats, the growth rate was reduced from 1.4–2.0 on the basal diet alone to 0.3–0.6 g. The normal growth rate was almost completely restored by the addition of *l*-cystine or *dl*-methionine (500 mg. per 100 g. of basal diet) to the diet.

F. A. R.

Behaviour of some Uramido-acids in the Nitrous Acid Method for the Determination of Amino-nitrogen. A. G. Gornall and A. Hunter. (*Biochem. J.*, 1940, 34, 192–197.)—The behaviour of 14 uramido-acids in Van Slyke's method for the estimation of amino-nitrogen was examined by measuring the amount of nitrogen evolved after 3, 30 and 60 minutes and 2½ hours. The uramido-group was decomposed by nitrous acid much more slowly than the α - and ω -amino groups in an amino-acid, but the rate of evolution of nitrogen varied greatly from acid to acid. The uramido-acids could be arranged in 3 distinct, if not sharply delimited, groups according to the amount of nitrogen evolved in 2½ hours. Those in Group I yielded distinctly less than 1 atom, those in Group II between 1.2 and 1.6, and those in Group III exactly, or almost exactly, 2. Certain generalisations could be drawn concerning the constitution of the acids in each group. Thus all the ω -uramido-acids examined belonged to the first group, the α -uramido-acids with straight chains (with one exception) belonged to the second, and the α -uramido-acids with branched chains (with one exception) belonged to the third. The exceptions— α -uramidopropionic acid and α -uramido-isohexoxic acid—yield less nitrogen than acids of analogous constitution.

F. A. R.

Histidine Detection and Estimation in Urine. E. Racker. (*Biochem. J.*, 1940, 34, 89–96.)—Histidine can be detected by the Pauly diazo reaction, but this is not specific. The Knoop bromine reaction is more distinctive, and has now been made quantitative by the elimination of three sources of error. First, phosphates are removed with baryta because their precipitation in the test inhibits the development of colour; secondly, the urine is decolorised with charcoal because its pigmentation interferes with the measurement of colour; thirdly, an excess of sodium urate is added to eliminate variations in the action of certain substances (including sodium urate) normally present in urine, which intensify the colour in the bromine reaction. Six ml. of urine are pipetted into a test-tube, 3 ml. of saturated baryta solution are added, and the

mixture is filtered. To the filtrate 3 ml. of *N* sulphuric acid and 100 mg. of decolorising charcoal are added, and the mixture is again filtered. Six ml. of the clear, colourless filtrate and 6 ml. of a standard solution of histidine hydrochloride (100 mg. in 1 litre of 0.1 *N* sulphuric acid) are treated with 2.5 ml. of a 5 per cent. suspension of sodium urate, and bromine in acetic acid solution (2 ml. in 100 ml. of glacial acetic acid diluted to 300 ml. with water) is then added in small portions until a faint yellow colour persists. After 10 minutes, with occasional shaking, the excess of bromine is removed by 1 or 2 drops of a saturated solution of arsenious oxide in 10 per cent. ammonia solution. The test-tubes are placed in boiling water for 5 to 7 minutes, and after a further 10 minutes the orange-red colours of the two solutions are compared in a colorimeter. The method will detect 1 part of histidine in 100,000 of urine, and the experimental error is about 10 per cent. with amounts between 10 and 20 mg. per 100 ml. After an examination of the behaviour of other substances, it was concluded that the test is not so distinctive as has previously been asserted. Some phenols (*e.g.* aminophenol), some alkaloids (*e.g.* morphine, tyrosine and tryptophane) react with bromine, but some of the colour can be extracted with amyl alcohol after addition of sodium carbonate; the colour due to histidine is not affected by this treatment. Histamine gives a colour that is much less intense and of a different shade.

F. A. R.

Method for the Estimation of Acetone Bodies in Blood, applicable also to the Estimation of Small Amounts of Mercury. L. A. Crandall. (*J. Biol. Chem.*, 1940, 133, 539–550.)—Into a 125-ml. Pyrex flask with a ground-glass joint are introduced 10 ml. of protein-free blood filtrate and 4 ml. of a modified Denigès reagent, prepared by dissolving 70 g. of mercuric sulphate in 6 *N* sulphuric acid and making up to 1 litre with 6 *N* sulphuric acid. The flask is fitted with a reflux condenser and the mixture is heated cautiously on a sand-bath, precautions being taken to avoid overheating. After a few minutes' boiling, 1.0 ml. of 5 per cent. potassium dichromate solution is added through the condenser and the heating is continued for a further 1½ hours. The solution is filtered through a sintered glass funnel while still warm, and the Denigès precipitate is thoroughly washed with water and then dissolved on the filter with 5 ml. of boiling conc. nitric acid, the solution being collected in a test-tube. The funnel is washed with water until the filtrate measures about 45 ml. Two ml. of 50 per cent. ferric nitrate solution are added, and the solution is diluted to 50 ml. with water. The concentration of mercury in this solution is estimated by adding 1 ml. of approximately 0.01 *N* potassium thiocyanate solution, and measuring the resulting colour in a photoelectric colorimeter. The mercury interferes with the colour formed when thiocyanate is added to an excess of ferric nitrate. The amount of mercury is calculated by

reference to a standard curve constructed with the aid of standard solutions of mercuric nitrate. The readings obtained in terms of mg. of mercury are converted into acetone bodies expressed as mg. of β -hydroxybutyric acid per 100 ml. of blood by multiplying by 15.4, or as mg. of acetone by multiplying by 6.33. The error of the method is less than ± 5 per cent. with amounts of acetone bodies ranging from 15 to 20 mg. per 100 ml. F. A. R.

The Glycogen of *Helix pomatia*. E. Baldwin and D. J. Bell. (*Biochem. J.*, 1940, **34**, 139-143.)—The hepatopancreas ("liver") of *Helix* is very similar in its metabolic behaviour to the liver of mammals. The glycogen isolated from the hepatopancreas after previous removal of the albumin glands (which contain galactogen), was found to contain no galactogen, contrary to the observations of previous workers. It had a specific rotation of $+192^\circ$, and its chain length was estimated at 11 to 12 units, so that it is apparently identical with the glycogen of rabbit liver, fish liver, horse muscle, etc., but different from the only other molluscan glycogen (*Mytilus edulis* glycogen) that has been examined, for which a value of 18 units was found. F. A. R.

Manometric Estimation of Nitrite in Solution and in Tissues. J. Brooks and J. Pace. (*Biochem. J.*, 1940, **34**, 260-267.)—Baumgarten and Margraff (*Ber.*, 1930, **63**, 1019) showed that nitrites can be estimated by measuring the amount of nitrogen liberated by the action of excess of amidosulphonic acid ($\text{NH}_2\text{SO}_3\text{H}$), since the rate of reaction is rapid compared with the rate of decomposition of nitrous acid. The reagent is prepared by bubbling sulphur dioxide through a saturated solution of hydroxylamine hydrochloride for 48 hours at room temperature and allowing the mixture to stand at -10°C . for a further 3 to 4 days. The crystals of amidosulphonic acid are recrystallised from hot (50°C .) water by cooling to -10°C . The amount of nitrogen evolved can be measured either in a Warburg apparatus or in a Van Slyke apparatus. When a Warburg apparatus is used, 1 ml. of the sodium nitrite solution is introduced into the main compartment of the vessel and 0.4 ml. of 1.7 per cent. amidosulphonic acid solution into the side bulb; only a slight excess of the acid is required, but the same results are obtained with more concentrated solutions. When thermal equilibrium at 25°C . is reached, the contents of the side bulb are tipped into the main compartment; the pressure increase reaches a steady value in less than 10 minutes. With the Van Slyke apparatus, 2 ml. of the nitrite solution, a drop of octyl alcohol and 4.75 ml. of water are drawn into the gas chamber, the mixture is shaken *in vacuo* for 5 minutes, and the liberated gas is expelled. The chamber is evacuated until the solution is in the lower fourth, and 0.25 ml. of saturated amidosulphonic acid solution are added through a mercury seal. The contents of the

chamber are shaken, with the mercury meniscus at the 50-ml. mark, for 5 minutes and any traces of carbon dioxide are absorbed in the usual way by the addition of 1 ml. of air-free *N* sodium hydroxide solution. The gas is brought to a volume of 0.5 ml. (or 2.0 ml.) and the pressure, p_1 , is read. The gas is ejected, the fluid meniscus is lowered to the 0.5 ml. (or 2.0 ml.) mark, and the pressure, p_2 , is read. Then the volume of nitrogen per 100 ml. is equal to $f(p_1 - p_2 - c)$ where f is a factor tabulated by Peters and Van Slyke (*"Quantitative Clinical Chemistry,"* 1932, p. 282) and c is a small correction obtained by a blank analysis in which 2 ml. of water are used in place of the sodium nitrite solution; $c = p_1 - p_2$. By either method, a concentration of 2×10^{-4} g. of sodium nitrite per ml. can be determined to within about 1 per cent. The quantitative evolution of nitrogen is not affected by the presence of sodium chloride, sodium nitrate, glucose, sucrose or tissue extractives. Nitrite in muscle can be estimated by the manometric analysis of an aqueous extract of the tissue, and, with certain limitations, by the direct action of amidosulphonic acid on tissue contained in the Warburg apparatus. F. A. R.

Enzyme Activity in Frozen Vegetables: Asparagus. M. A. Joslyn and C. L. Bedford. (*Ind. Eng. Chem.*, 1940, **32**, 702-706.)—Samples of asparagus, cut in 1933, 1937 and 1938, had been blanched in water for different periods of time ranging from 1 to 20 minutes, and at various temperatures, and had then been packed in tins, which were sealed and stored at -17°C . The samples were all examined for catalase, peroxidase and ascorbic acid oxidase activity and aldehyde-content by methods already described in earlier publications (*Ind. Eng. Chem.*, 1936, **28**, 595; 1938, **30**, 1068; 1939, **31**, 751), the 1933 series after storage for 4.5 years, the 1937 after 15 months, and the 1938 after 3 months. In the 1933 series, maximum catalase activity was observed in the samples packed at 40°C ., and maximum acetaldehyde-content in the samples packed at 60°C .; there was a fairly close correlation between the acetaldehyde-content and the catalase activity. Inactivation of catalase was most rapid above 75°C . The peroxidase activity of these samples, when measured in terms of pyrogallol-content, did not run parallel with catalase activity, though this also fell off markedly above 75°C . Asparagus packed in 1937 and 1938 had greater peroxidase activity, as measured colorimetrically with both guaiacum and benzidine, than the 1933 samples. The enzyme responsible for the guaiacum reaction was more thermolabile than that responsible for the benzidine reaction. The ascorbic acid oxidase activity of asparagus tissue was small. The presence of a weak oxidase, readily destroyed by heating, was established. The asparagus packed in 1933 was also examined periodically for colour and flavour. At first the flavour was good, but off-flavours developed

on storage, and after 4½ years all the samples had distinctly unpleasant flavours. Scalding in steam or boiling water for 2 to 5 minutes produced the most satisfactory flavour. The gum guaiac peroxidase reaction appeared to be more closely correlated with flavour retention than the other enzyme reactions. A similar correlation was observed with the 1937 and 1938 packs, and asparagus scalded for 3 minutes in boiling water retained its original flavour on storage for 15 months. Other factors beside enzyme inactivation appear to be involved in flavour retention, such as the length of time between cutting and processing.

F. A. R.

Carbonate Veronal Buffer Solution covering pH 7.5 to 10.7. E. J. King and G. E. Delory. (*Enzymologica*, 1940, 8, 278-279.)—Most buffers are unsatisfactory for the study of the hydrolysis of phosphoric esters by mammalian phosphatases. For this, a combined Michaelis-Kolthoff buffer solution is proposed, which covers pH 6.8 to 9.6. It is prepared as follows:—25 ml. of *M*/10 sodium veronal are carefully pipetted on to *x* ml. of *N*/10 hydrochloric acid in a 100-ml. flask. After addition of 25 ml. of *N*/10 sodium carbonate solution and gently mixing, the solution is diluted to 100 ml. The following table gives the pH values of different mixtures. The determinations were made with the hydrogen electrode and the saturated calomel electrode, and the standard of reference was a sodium acetate and acetic acid buffer of pH 4.63 (*cf.* Sendroy, *Trans. Electrochem. Soc.*, 1934, 74, 595).

| <i>x</i> | pH at 22° C. | pH at 37° C. |
|----------|--------------|--------------|
| 2.5 | 10.7 | 10.4 |
| 7.5 | 10.3 | 10.1 |
| 12.5 | 9.95 | 9.79 |
| 20.0 | 9.44 | 9.27 |
| 25.0 | 9.00 | 8.83 |
| 30.0 | 8.44 | 8.34 |
| 40.0 | 7.96 | 7.87 |
| 50.0 | 7.48 | 7.45 |

When kept in well-closed, paraffin-waxed bottles, these mixtures showed no change in pH values for at least 10 months. E. B. D.

Use of Ascorbic Acid as a Substrate in Oxidase Measurements. B. D. Egell and F. Gerhardt. (*J. Agric. Res.*, 1940, 60, 89-99.)—A study of the oxidative enzymes of fruit is of importance for an understanding of the changes that occur in stored fruit, and for this reason a more sensitive method of measuring small changes in enzyme activity is desirable. Guthrie (*J. Amer. Chem. Soc.*, 1930, 52, 3614; *cf.* ANALYST, 1930, 55, 709) determined oxidase activity iodimetrically, using as substrate a solution of glucose heated with dilute sodium hydroxide solution. Such

a substrate has many disadvantages: (1) its composition is uncertain and variable, (2) it is usually coloured, and (3) it contains substances injurious to the enzyme. The use of ascorbic acid as substrate in place of the heated glucose solution has been investigated. Ascorbic acid has been found to be free from the disadvantages cited, and in addition it is readily oxidised, enabling smaller differences in enzyme activity to be detected. Twenty-five ml. of a 0.1 to 0.4 per cent. solution of ascorbic acid are introduced into a Van Slyke and Cullen aeration tube, and 1 to 10 ml. of the juice or extract containing the enzyme is added. A similar tube is filled with the substrate and a sample of the boiled juice or extract to serve as a blank. Air is drawn for 1 hour through the tubes, which are maintained at 25° C., and after aeration the solutions are transferred with 50 ml. of water to flasks containing 25 ml. of 10 per cent. trichloroacetic acid solution. About 20 ml. of 0.1 *N* potassium iodide solution are added, and after 30 minutes the solutions are titrated with 0.01 *N* sodium thiosulphate solution. The difference in the titration values of the boiled and unboiled samples is a measure of the oxidase activity. Preliminary experiments indicated that the optimal pH value for oxidase activity was approximately that of the original extract, *e.g.* 4.0 for apple juice and 6.2 for potato juice. Differences in the concentration of the substrate from 0.1 to 0.4 per cent. had little effect on the enzyme activity. The oxidation of ascorbic acid was certainly enzymic, the possibility of catalytic oxidation by traces of copper being excluded.

F. A. R.

Vitamin A Destruction in Fish-liver Oils. E. J. Simons, L. O. Buxton and H. B. Colman. (*Ind. Eng. Chem.*, 1940, 32, 706-708.)—Several different kinds of fish-liver oils were stored in the dark in small tubes open to the air at a temperature of $34.5 \pm 0.5^\circ \text{C.}$, and the peroxide values and vitamin A contents were determined at intervals. The peroxide values were measured by the methods of Wheeler* (*Oil and Soap*, 1932, 9, 89), and the vitamin A contents spectrophotometrically, a factor of 2000 being used for converting $E_{1\text{cm}}^{1\%}$ 328mμ into I.U. per g. When the percentage of vitamin A destroyed at any given time was plotted against the peroxide value at that time, the resulting curves fell into two distinct groups. With the more unsaturated oils, such as cod, pollack and the U.S.P. reference oil, the percentage of vitamin A oxidised was smaller at various peroxide values than in the less unsaturated oils, such as dogfish, halibut and swordfish liver oils, at similar peroxide values. Within each of these two groups, the percentage of vitamin A oxidised was related to the peroxide value of the oil, and the higher the peroxide value the more vitamin

* One-gram samples of oil are dissolved in 25 ml. of a mixture (3 : 2) of glacial acetic acid and chloroform, 0.5 ml. of saturated potassium iodide solution is added, and the mixture is mixed gently for one minute. Fifty ml. of water are added, and the liberated iodine is titrated immediately with 0.02 *N* sodium thiosulphate solution, starch being used as indicator.

A was oxidised. The results are explained by assuming that two reactions occur, the first being the formation of peroxides and the second interaction between the peroxides and the vitamin. The ratio of rate of peroxide formation to rate of oxidation of vitamin A, is different for each of the two groups of oils.

F. A. R.

Vitamin A Activity and Vitamin B₁ Content of Soya-beans and Cowpeas. J. O. Halverson and F. W. Sherwood. (*J. Agric. Res.*, 1940, 60, 141-144.)—Nine varieties of soya-bean (*Soja max.*) and eight of cowpeas (*Vigna sinensis*) were assayed biologically for vitamin A activity and vitamin B₁ content. None of the samples contained appreciable amounts of vitamin A. The soya-beans contained 3.2 to 4.8 I.U. of vitamin B₁ per g., with an average value of 3.8, and the cowpeas 2.3 to 3.7, with an average of 3.0. F. A. R.

Observations on the Distribution of Vitamin B₁ in some Plant Families. M. Pyke. (*Biochem. J.*, 1940, 34, 330-334.)—In continuation of work already published (*cf. ANALYST*, 1940, 65, 180), a survey has been made, by the method previously described, of a large variety of fruit and vegetables. The following are the most important of the additional results now reported, the vitamin B₁ content being expressed in I.U. per 100 g. I. *Leaves and tops*.—New Zealand spinach, seakale, celery, 15 to 25. II. *Roots, etc.*—Beetroot, onions, 6 to 8; turnip, radish, 11 to 13; chicory, swede, 25; potato, parsnip, 30 to 38; leek, Jerusalem artichoke, 50 to 76. III. *Seeds and nuts*.—Butter-beans, 140; Barcelona nuts, 38; cob nuts, 76; Brazil nuts, 340. IV. *Fruit*.—Pineapple, cucumber, pear, peach, blackberry, lemon, currant, grape, 6 to 20; orange, 34; damson, 38; plum, green-gage, 66. It is concluded that in general the vitamin B₁ content is highest in seeds, and that the amount in leaves is relatively constant irrespective of the botanical family. F. A. R.

Vitamin C Content of Honey. E. Becker and R. F. Kartos. (*Z. Unters. Lebensm.*, 1939, 78, 305-308.)—The reducing substances in 12 samples of honey determined by titration with N/100 iodine solution, ranged from 3.5 to 89 mg. per 100 g. calculated as ascorbic acid. The amounts found in 4 samples by titration with 2,6-dichlorophenolindophenol ranged from 5.6 to 32.5 mg. Biological tests on guinea pigs, however, did not show any antiscorbutic action, and the authors therefore concluded that the honeys examined did not contain any vitamin C, but that an unknown reducing substance was present. D. A.

Bacteriological

Efficiency of the Wells Air Centrifuge as Determined by Air-washing Technique. K. MacDonald. (*Amer. J. Hyg.*, 1940, 31, 85-87.)—The Wells Air Centrifuge technique has been accepted for the bacteriological analysis of

air by most investigators in America and has been adopted by the American Public Health Association. It is not claimed by Wells that the instrument takes out all the bacteria, nor that it takes out the same proportion in every instance, and it was thought desirable to ascertain its average efficiency. The Palmer water spray sampler, which by means of an electrically driven fan aspirates air through a fountain spray curtain of water, retains 98 per cent., by actual test, of small particles of the same size as bacteria and spores. Parallel experiments were made with the Palmer apparatus and the Wells Air Centrifuge on air of an experimental room sprayed with 25 to 90 ml. of a 48-hour broth culture of *S. marcescens* per 10 cubic feet. Thirty minutes after the spraying of the air samples were taken, and again after every 15 minutes, until a series of 5 was completed, each sample consisting of 10 cubic feet of air and the sampling time being 5½ minutes. Thirty series of tests were made. Following each series the saline wash was plated out in agar and incubated, together with the contents of the corresponding Wells tube, at room temperature for 4 days. The results showed that the Wells Air Centrifuge has an efficiency of 96 per cent. in removing artificially inoculated droplet nuclei, but as the concentration of normal air-borne organisms increases, the average efficiency falls to 75 per cent. D. R. W.

Colouring Matters of *Penicillium carmino-violaceum* Blourge, with a Note on the Production of Ergosterol by this Mould. H. G. Hind. (*Biochem. J.*, 1940, 34, 67-72.)—Cultures of *P. carmino-violaceum* grown on carbohydrate media are characterised by a colouring of the medium, yellow when acid and claret red when alkaline. The pigments responsible for these colour changes have now been isolated, and are given the names carviolacin and carviolin. The composition of carviolacin corresponds with the formula C₂₀H₁₆O₇, and that of carviolin with the formula C₁₈H₁₄O₆; both are monomethyl ethers of complex anthraquinone derivatives. Ergosterol was isolated from the mould mycelium. F. A. R.

Constitution of Carviolin: a Colouring Matter of *Penicillium carmino-violaceum* Blourge. H. G. Hind. (*Biochem. J.*, 1940, 34, 577-579.)—Carviolin trimethyl ether was shown to be identical with the product obtained by methylating ω -hydroxyemodin, and carviolin C₁₈H₁₄O₆, which had previously been shown to contain one methoxy group, is therefore a monomethyl ether of ω -hydroxyemodin; the compound probably has a free β -hydroxyl group. F. A. R.

Studies on the Biochemistry of Micro-organisms. Emodic Acid and ω -Hydroxyemodin, Metabolic Products of *Penicillium cycloptum* Westling. W. K. Anslow, J. Breen and H. Raistrick. (*Biochem. J.*, 1940, 34, 159-168.)—The mycelium of a strain of

P. cyclopium yielded two derivatives of 2-methyl-anthraquinone, namely, emodic acid ($C_{14}H_8O_7$) in the form of orange needles, m.p. 363° to 365° , and ω -hydroxy-emodin ($C_{14}H_{10}O_6$), also as orange needles, m.p. 288° C. Emodic acid is 4 : 5 : 7-trihydroxyanthraquinone-2-carboxylic acid and ω -hydroxy-emodin is 4 : 5 : 7-trihydroxy-2-(hydroxymethyl)-anthraquinone. *Frangula*-emodin (4 : 5 : 7-trihydroxy-2-methylanthraquinone), ω -hydroxy-emodin and emodic acid, thus bear the same relationship to one another as do chrysophanic acid, aloë-emodin and rhein.

F. A. R.

Toxicological and Forensic

Detection of Chloralose in Urine. M. P. Cheramy. (*J. Pharm. Chim.*, 1940, [9], 1, 233-234.)—Preparations of chloralose sold under names resembling those of the barbituric hypnotics have been used in attempted suicide. As the resulting coma resembles that produced by the barbiturates, large doses of strychnine are administered, and in one instance this treatment resulted in violent convulsions. Examination of the urine failed to reveal the presence of barbituric compounds, and after recovery the patient stated that he had taken a quantity of "somenal" corresponding with 2.25 g. of chloralose. A simple test for the presence of chloralose in urine is therefore required. The reaction described by Ross (*J. Biol. Chem.*, 1923, 58, 641; Abstr., ANALYST, 1924, 49, 144), although neither delicate nor specific, serves for the detection of the moderately large amounts of chloralose that would be present in the urine of those attempting suicide with this drug. The test may be applied directly to the urine, but the following procedure is preferable:—The urine (20 ml.) is treated with 1 ml. of conc. sulphuric acid and 0.5 g. of activated carbon, and the mixture is boiled beneath a reflux condenser for 5 to 10 minutes. The cooled and filtered liquid is heated in a boiling water-bath with 2 or 3 ml. of pyridine and an equal volume of 30 per cent. sodium hydroxide solution for 1 or 2 minutes. In the presence of chloralose the pyridine layer is coloured rose to cherry-red, and the reaction will detect a concentration of 0.1 g. per litre. The reaction does not distinguish chloralose from other tri-halogenated products of methane (chloroform, chloral, bromoform, iodoform), but renders possible the rapid recognition of the type of hypnotic that has been taken.

A. O. J.

Estimation of Barbiturates in Blood. G. A. Levy. (*Biochem. J.*, 1940, 34, 73-77.) None of the methods given in the literature for the estimation of the more stable barbituric acids can be applied to unstable barbiturates, such as evipan and pentothal. These compounds are insoluble in acid and neutral solution, and unstable in alkaline solution, rendering impossible the usual methods of extraction; they can, however, be isolated by

drying the blood with anhydrous sodium sulphate and extracting the powder in a Soxhlet apparatus with a fat-solvent. After purification, the barbiturate is estimated by Koppányi's colour reaction with cobalt acetate and isopropylamine in chloroform solution (Dille and Koppányi, *J. Amer. Pharm. Ass.*, 1934, 23, 1079). The method is applicable to the estimation of other barbiturates. For quantities of 1 mg. or more, the observed maximum error is 20 per cent., and for smaller amounts, the error is correspondingly greater. In a mortar (250 ml. capacity), 20 ml. of blood (coagulated or citrated) are mixed with 2 g. of sodium dihydrogen phosphate, and 40 g. of anhydrous sodium sulphate are added in small portions with constant grinding. The mortar is placed in a desiccator for 10 to 15 minutes to allow of complete drying, and the cake is then broken up and packed into a 100-ml. Soxhlet thimble. The solid is extracted for 3 hours at 50° C. with 80 ml. of a mixture (1 : 1) of petroleum spirit (b.p. 30° to 40° C.) and peroxide-free ether. At the end of the extraction the contents of the flask are shaken for a few minutes with 0.25 g. of a mixture (3 : 1) of charcoal (Merck's "Ultracarbon" washed with ether containing a few drops of glacial acetic acid, and then with ether) and magnesium oxide. The suspension is filtered, and the filtrate is distilled. The crystals of barbiturate that remain in the flask are taken up in a little chloroform and transferred to a 10-ml. graduated centrifuge tube and the solution is made up to a suitable volume. To 2 ml. of the solution are added 0.1 ml. of a 1 per cent. solution of cobalt acetate in methyl alcohol and 0.6 ml. of a 5 per cent. isopropylamine solution in methyl alcohol; the colour is compared with that given by 2 ml. of a standard solution similarly treated.

F. A. R.

Estimation of Carbon Monoxide in Blood. C. Scholten. (*D. Z. gericht. Med.*, 1939, 30, 292-296; *Z. Unters. Lebensm.*, 1940, 79, 296.)—An ammoniacal solution of silver oxide containing pyridine is mixed in a tube with a measured volume of fresh unclotted blood, and the tube is closed with a rubber stopper and left for 10 minutes in a water-bath at 70° C. The depth of the brown colour formed by reduction of the silver salt to colloidal silver is measured in a colorimeter. The degree of accuracy is about 5 per cent.

E. M. P.

Lead-content of Human Hair. K. N. Bagchi, H. D. Ganguly and J. N. Sirdar. (*Ind. J. Med. Res.*, 1940, 27, 777-791.)—About 200 samples of hair from apparently healthy Europeans, Anglo-Indians and Indians of either sex and of various ages were examined. Contamination by extraneous lead compounds was eliminated by washing the samples, in bulk, successively with hot soap solution, ether, hot dilute sodium hydroxide solution, water, alcohol and ether. Attempts to reduce the number of stages were unsuccessful,

although further treatment (with alkali, acid, water, ammonium acetate solution, alcohol and ether, in succession) resulted in no further reduction in the lead content. The lead was determined by the dithizone method of Lynch, Slater and Osler (ANALYST, 1934, 59, 787; 1935, 60, 32), slightly modified, 2 g. of hair usually being taken. The values found varied widely, namely, from 3.0 to 508 p.p.m., and since women's black hair contained the largest amounts (170 to 508 p.p.m.), men, and women's grey hair the least (3.0 to 21.0 p.p.m.) and brown, auburn and other shades of hair intermediate quantities (9.0 to 16.5 p.p.m.), it is suggested that lead is a factor in the production of the characteristic hair pigmentation of different nationalities. The Bengalee Hindu married women use a cheap grade of "vermilion" (consisting of red lead mixed with a red synthetic dye and scented) in order to mark the thin line, about 1 inch long on the anterior end of the hair-parting, which is indicative of unwidowed married life, and the average lead-content of their hair (180 p.p.m.) is much higher than that of the Bengalee Mohammedan women (50.4 p.p.m.), who do not use it. The hair of children contains relatively less lead than that of their parents, unless there is evidence of abnormal exposure; girls seem to be more susceptible to such exposure, and absorb larger quantities of lead than boys. Thus in one family the hair of the son (7 years old) contained 9 p.p.m., and that of the daughter (3½ years old) 75 p.p.m. of lead. There is evidence that the use of vegetable hair oils (e.g. sesame oil) aids absorption of lead through the scalp in the same way that greasy cosmetics containing lead are absorbed through the skin (cf. Monier-Williams, "Lead in Food," 1938, p. 2; ANALYST, 1939, 64, 32). The values found for the male population of the various Indian provinces, except Bengal, (20.0 to 22.7 p.p.m.) are of the same magnitude as that found for the average European of either sex, including Jews and Anglo-Indians (18.4 to 20.8 p.p.m.), and the hairs of European residents in Calcutta were similar in average lead-content. Hair from Bengalee Mohammedan males, however, gave a much higher figure (42.4 p.p.m.), although no reason for this has yet been found. Relatively large quantities of lead were found in the urine and faeces of some Hindu men and women and, since correspondingly high quantities were then found in the hair, it is considered that lead is absorbed into the general circulation and eliminated through the hair. Thus 0.04, 387.1 and 241.0 p.p.m. of lead were found in the urine, faeces and hair of a worker in a lead factory. In many instances a mild form of alopecia was associated with high lead-contents in the hair, and it is suggested that chronic lead intoxication may prevent access of nourishment to the follicles, with the result that the hair falls out. Lead is not always distributed uniformly in a hair; in one instance (female) the distal end contained 66 mg. and the proximal end 48 mg., whilst in another there was little difference between the ends.

This was to be expected, since the excretion of lead through the hair varies from day to day, and it may prove of medico-legal importance, e.g. as an indication of the period of ingestion of a poison. The form in which the lead is present in hair is not yet known, but a combination with phosphorus is suspected. Other metals (e.g. cobalt, copper, zinc, and manganese) also occur in hair in relatively large quantities, and it is believed that the surplus metals which have served their purpose (e.g. as catalysts in the physiological system) are removed in this way (cf. ANALYST, 1939, 64, 698). J. G.

Examination of Dusts from Lungs Produced by Mining and other Processes. D. G. Beadle. (*J. Chem. Met. Mining Soc. S. Africa*, 1940, 40, 286-287.)—Methods of measurement of the size distributions of dust from silicotic lungs are discussed (cf. H. S. Patterson, *id.*, 1939, 39, 229). In Jones's method (*J. Hyg.*, 1933, 33, 309; and ANALYST, 1934, 59, 124, 191) the lung is disintegrated with nitric acid, the resulting slime is filtered, and the residue is dried and ignited; the resulting deposit, however, is lumpy (probably because of the presence of phosphates which form glassy compounds on ignition), and is therefore unsuitable for size-distribution tests. It is preferable to use Fox's method, in which the tissue is boiled in a Kjeldahl flask with a mixture of nitric and sulphuric acids containing a crystal of copper sulphate. When the organic matter has been destroyed the residual siliceous dust may be separated by filtration and suspended in water, a drop of the suspension is allowed to evaporate on a microscope cover-glass, and the residual deposit is ignited and examined under the microscope. It was found that 22.5, 42.5 and 35.0 per cent. of the particles were less than 0.2, 0.2 to 0.8 and 1.2 to over 5.0 microns in size, respectively. In Simson's method the minced lung tissue is digested with papain in an incubator for 10 days at 54° C., the resulting mixture is diluted and centrifuged, and the deposit is washed by a combination of centrifuging and decantation. Phosphates are then removed by warming with hydrochloric acid for 30 mins. and the residue is again washed as described and mounted for examination. It was found that 48.1, 29.2, 5.2 and 17.5 per cent. of the particles were less than 0.2, 0.2 to 0.4, 0.8 and 1.2 to over 5.0 microns in size, respectively. The difference between the two methods probably arises from a loss of fine particles during filtration in the Fox method. J. G.

Active Principles of Leguminous Fish-poison Plants. IV. Isolation of Malaccol from *Derris malaccensis*. S. H. Harper. (*J. Chem. Soc.*, 1940, 309-314.)—The ethereal extract of the root of *Derris malaccensis* gives a gelatinous deposit (about 1.5 per cent. on the root). The substance is present in the root in a stabilised non-crystalline form, but when purified by crystallisation from benzene or

ethyl acetate yields bright yellow prisms or needles, m.p. 225°C. , re-melting at 244°C. (the double m.p. is due to racemisation), and having $[\alpha]_{\text{D}}^{18} = +190^{\circ}$ in chloroform, $+67^{\circ}$ in benzene. It was found to be identical with a substance isolated by Meyer and Koolhas from Sumatra derris root (*Rec. Trav. chim. Pays-Bas*, 1939, 58, 207; *Abst., ANALYST*, 1939, 64, 295), and was called "malaccol." Analytical data agreed with the formula $\text{C}_{20}\text{H}_{32}\text{O}_7$; this formula bears the same relationship to that of elliptone as does the formula of sumatrol to that of rotenone, and malaccol is thus isomeric with elliptolone (*J. Chem. Soc.*, 1939, 1424). Malaccol differs from other members of the group in giving a $+$ rotation in benzene solution, but the introduction of a phenolic OH in position 15 in each instance gives a positive increment to the specific rotation, and malaccol is regarded as thus fitting into the *l*-series (rotenone—deguelin—elliptone—sumatrol—toxinol—malaccol). On maintaining the temperature of *l*-malaccol just above the lower m.p. a rapid loss of optical activity is observed and *l*-malaccol readily sublimates at 195°C. in a high vacuum to give a 75 per cent. racemised product. D. G. H.

Gas Analysis

Determination of Methylpropene by Means of a Modified Denigès Reagent. A. Newton and E. J. Buckler. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 251-254.)—An investigation of the reaction between methylpropene and Denigès' reagent (*Compt. rend.*, 1898, 126, 1043) showed that the variation in the composition of the precipitate is due partly to the excessive acidity caused by the formation of nitric acid in the reaction and partly to the replacement of the nitrate radical by hydroxyl (or possibly water) during the washing of the precipitate. When the reagent is modified by addition of sufficient sodium hydroxide to convert the mercuric nitrate into the basic salt the total weight of the precipitate cannot be taken as a measure of the methylpropene absorbed, but the amount of mercury in the precipitate is equivalent to the methylpropene. The modified reagent is prepared as follows:—Mercuric oxide (100 g.) is mixed to a paste with 100 ml. of water and dissolved in the minimum amount (about 90 ml.) of 70 per cent. nitric acid. The solution is treated, drop by drop, with a solution of 10 g. of sodium hydroxide in about 15 ml. of water until a slight permanent precipitate of basic mercuric nitrate is formed. The mixture is then diluted with water to 2 litres, filtered and stored in a dark glass-stoppered bottle. The apparatus required for the determination consists essentially of a water-jacketed gas burette with a compensator and an absorption tube. The sample of gas, varying from 35 ml. for 20 per cent. methylpropene to 150 ml. for 1 per cent. methylpropene, is introduced into the burette and, after the lapse of 30 seconds, its volume

and pressure are measured. The reagent (50 ml.) is drawn into the absorption tube which is then evacuated by means of a water-pump, closed and attached to the burette. The sample of gas is transferred to the absorption tube, which is then detached and shaken vigorously for 5 minutes and, after the admission of air, for a further 2 minutes. The liquid is run out from the absorption tube, which is rinsed, and the combined absorbent and washings are heated gently to 100°C. (but not allowed to boil). When cold the liquid is filtered through a No. 3 (or preferably No. 4) sintered glass crucible, and the precipitate is washed with cold water until the filtrate gives no precipitate with ammonium sulphide. The precipitate is dissolved by warming with about 10 ml. of 70 per cent. nitric acid, and the solution is oxidised by the addition of a saturated potassium permanganate solution until the pink colour persists. The excess of permanganate is destroyed by the addition of a slight excess of a 10 per cent. oxalic acid solution and, after dilution to 100 to 150 ml. and addition of 2 ml. of a saturated solution of ferric alum in 50 per cent. nitric acid, the liquid is titrated with 0.1 *N* potassium thiocyanate solution. Each ml. of 0.1 *N* potassium thiocyanate solution is equivalent to 0.01003 g. of mercury or 0.1601 ml. of methylpropene (at 0°C. and 760 mm.). Good results are obtained by the use of a volume of reagent equal to about 10 times the volume of methylpropene to be absorbed. Ethene, propene and 1, 3-butadiene were slowly absorbed by the reagent, but gave no precipitate when the solution was boiled. A mixture of 1- and 2-butenes prepared by the dehydration of secondary butanol was slowly absorbed, and yielded a faint turbidity corresponding with 0.37 per cent. of methylpropene in the gas. When the gas was shaken twice with 67 per cent. sulphuric acid the residue was slowly absorbed by the reagent but yielded no precipitate. Hurd and Goldsby (*J. Amer. Chem. Soc.*, 1934, 56, 1812) report that 2-methyl-2-butene gives no precipitate with the acid Denigès reagent. With the modified reagent the mixture of pentenes obtained by dehydrating 2-methyl-2-butanol gave a heavy precipitate, and a C_6 fraction obtained from debutanised cracked gasoline behaved similarly. This was probably due to the presence of 2-methyl-1-butene in the samples. The possibility of applying the method to the determination of pentenes is being studied; 1- and 2-butenes neither give a precipitate with the reagent nor interfere with the determination of methylpropene. The method is entirely satisfactory in the presence of C_2 , C_3 and C_4 olefines, but pentenes must be absent. A. O. J.

Organic

Determination of Lactic and Pyruvic Acids by means of Periodic Acid. (Mile) R. Boisson. (*J. Pharm. Chim.*, 1940, [8], 1, 240-255.)—The behaviour of lactic and pyruvic

acids on oxidation with periodic acid (Boisson and Fleury, *J. Pharm. Chim.*, 1939, 30, 145, 307) affords means for their analytical determination. It has been shown (*loc. cit.*) that when dilute solutions of lactic acid are oxidised at 100° C. with periodic acid, acetaldehyde and carbon dioxide are formed. The aldehyde, which must be removed as soon as it is formed, is passed by means of a current of air into alkaline iodomercurate solution, and the precipitated mercury is determined by a volumetric method. Ten ml. of an aqueous solution, containing 0.1 to 1.0 per cent. of lactic acid, are mixed with 10 ml. of 10 per cent. sodium periodate solution and 2 ml. of 10*N* sulphuric acid and heated in a flask provided with the means of leading an air current at the rate of 10 to 20 litres per hour through the liquid. The vapour is led through a reflux water-condenser into two absorption flasks in series containing the iodomercurate solution. Concentrated iodomercurate solution is prepared by dissolving 27 g. of mercuric chloride and 72 g. of potassium iodide in sufficient water to produce 250 ml. Ten ml. of this solution are mixed with 15 ml. of sodium hydroxide solution (sp.gr. 1.33), and 5 ml. of this mixture are placed in the second absorption flask and the remainder, with 5 ml. of a 20 per cent. suspension of barium sulphate in water, in the first. Distillation is allowed to proceed for 30 minutes. The combined liquids in the absorption flasks are acidified with dilute sulphuric acid, a known excess of *N*/10 iodine solution is added, and the mixture is allowed to stand with frequent shaking until all the precipitated mercury has dissolved. The residual iodine is then titrated with *N*/10 sodium thiosulphate solution. The factor to convert ml. of *N*/10 iodine solution into mg. of lactic acid is 4.5. With the following modifications the method may be used as a micro-method. Five ml. of lactic acid solution containing 0.5 to 1 mg. of lactic acid, 3 ml. of 10 per cent. sodium periodate solution and 1 ml. of 10*N* sulphuric acid are placed in the distillation flask. The first absorption flask contains 6 ml., and the second 1 ml., of alkaline iodomercurate solution. During the first 15 minutes water is not circulated through the condenser, and the volume of the reaction mixture is thereby reduced to about one-half. Water is then circulated, and the distillation is continued for a further 45 minutes, the air-current being maintained. The contents of the absorption flasks are acidified with 5 or 6 ml. of 10*N* sulphuric acid, and the determination is carried out as previously described, 0.02*N* iodine solution and 0.01*N* sodium thiosulphate solution being used. A control determination is made with 6 ml. of the iodomercurate solution. The application of the method to the determination of lactic acid in presence of glucose was studied. Methods depending upon the removal of glucose by chemical means were found unsuitable, and the method finally adopted was extraction of the lactic acid with ether from an aqueous solution saturated with ammonium sulphate. A simple form of

extractor for this purpose is described. The lactic acid is then extracted from the ethereal solution with dilute alkali solution, and this extract is submitted to the process already described.

For the determination of pyruvic acid two methods are available. The solution containing 5 to 30 mg. of pyruvic acid is heated beneath a reflux air-condenser for one hour with 5 ml. of 0.1*M* sodium periodate solution in 30 per cent. (v/v) sulphuric acid. With 5 to 15 mg. of pyruvic acid the time may be reduced to 30 minutes. To the cooled mixture excess of sodium bicarbonate, 15 ml. of 0.1*M* arsenious anhydride solution and 1 ml. of 20 per cent. potassium iodide solution are added (*cf. Abst., ANALYST*, 1933, 58, 307). After 10 minutes the residual arsenious anhydride is titrated with *N*/10 iodine solution. The result of a blank determination is deducted. The factor to convert ml. *N*/10 into mg. of pyruvic acid is 8.8. By the second method the acidity to thymolphthalein indicator is determined before and after the oxidation. Since pyruvic acid is oxidised to the equivalent amounts of acetic acid and carbon dioxide, the diminution of acidity to this indicator represents the amount of periodic acid reduced. The conditions of concentration and heating are the same as for the first method. The cooled liquid is titrated to the pale-blue end-point of the indicator with *N*/10 sodium hydroxide solution. The control determination is made with 5 ml. of sodium periodate solution and 5 ml. of the solution to be examined. The method is rapid, but less precise than the first. It was found impossible to determine lactic acid in mixtures of lactic and pyruvic acids by the methods described unless the molecular concentration of the lactic acid was at least equal to that of the pyruvic acid.

A. O. J.

Identification of 2-Aminoethanol. B. Kelsor. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 284.)—In aqueous solution 2-aminoethanol (monoethanolamine) can be identified readily by its reaction with phthalic acid to form 2-hydroxyethylphthalimide (m.p. 127° C.). The solution of 2-aminoethanol is treated with an equivalent amount of phthalic acid dissolved in hot water. The mixture is evaporated to dryness, and the residue is heated at 210° C. for 5 minutes. The phthalimide, which is formed quantitatively, is recrystallised from water, or, if inorganic impurities are present, from an anhydrous solvent such as benzene or absolute alcohol, and its m.p. is determined. Conversely, the reaction may be used for the identification of phthalic acid. The concentration of 2-aminoethanol in the sample is assumed, or determined by titration with acid, each ml. of *N* hydrochloric acid being approximately equivalent to 0.061 g. of the amine. 2-Aminoethanol can also be identified in aqueous solution by formation of its oxalate (m.p., 199° to 200° C. uncorr.; corr., + 3.8° C.). In addition, this salt may be

converted into *N,N'*-bis-(2-hydroxyethyl)-oxamide, which melts at 168° C. uncorr. (corr., + 2.4° C.). The solution of 2-aminoethanol is treated with an equivalent amount of an oxalic acid solution and evaporated to dryness. The residue is heated at 110° C. for 5 minutes and recrystallised from 70 per cent. alcohol and its m.p. is determined. On heating to about 222° C. brisk boiling occurs, with formation of the oxamide. The reaction may be used for the identification of oxalic acid.

A. O. J.

Determination of Organic Peroxides.

H. A. Liebhafsky and W. H. Sharkey. (*J. Amer. Chem. Soc.*, 1940, **62**, 190-192.)—Organic peroxides in *n*-butyl ether may be determined by iodimetric titration in glacial acetic acid. Satisfactory blank tests cannot be made by this method, but the need for a blank test is avoided by adding sodium bicarbonate to the reaction mixture to drive out air. To 25 ml. of glacial acetic acid and approximately 1.5 g. of sodium bicarbonate in a glass-stoppered flask, 1 ml. of potassium iodide solution (0.4 g. per ml.) is added. The faintly yellow mixture is titrated with *N*/100 thiosulphate solution, added two drops at a time, until the colour is the faintest perceptible to the eye. (Starch is an unsatisfactory indicator here.) As the reaction in glacial acetic acid does not appear to be instantaneous, the successive additions of thiosulphate should be made at intervals of about 10 seconds. The sample for analysis (usually 5 ml. of *n*-butyl ether) is then added, the flask is stoppered and kept in the dark for 5 minutes, and the mixture is titrated with *N*/100 thiosulphate solution. If, after 5 minutes more in the dark, no further liberation of iodine occurs, the titration is satisfactory. Experiments with test solutions showed the reaction to be complete in less than ten minutes, and accurate to within two drops of *N*/100 thiosulphate solution. A study of the formation and decomposition of the peroxides in *n*-butyl ether has shown that they are increased by exposure to ultra-violet radiation or to daylight, and still more rapidly by bubbling oxygen through the boiling ether, whilst they are destroyed by heating under reflux. Hydrogen peroxide or benzoyl peroxide in glacial acetic acid may also be determined accurately by this method.

E. B. D.

polysaccharide with $[\alpha]_D^{20} = -51.2^\circ$. This is composed of 1 monomethylhexuronic acid residue and 6 xylose residues. F. A. R.

Examination of Damaged Cotton by the Congo Red Test: Further Developments and Applications. G. G. Clegg.

(*J. Textile Inst.*, 1940, **31**, T. 49-68.)—Slight modifications of the original test (*cf.* Bright, *ANALYST*, 1926, **51**, 593) are described. The sample is (1) immersed for 3 minutes in the sodium hydroxide solution, which should contain a wetting-agent (*e.g.* 0.5 per cent. Shirlacrol); (2) washed in water; (3) placed in the Congo red reagent for 10 minutes; (4) washed; (5) immersed in 18 per cent. sodium hydroxide solution. The concentration of alkali used in stage (1) must be selected so as to produce a controlled degree of swelling; thus, if the fibres are already damaged so that the cuticle is ruptured and the secondary cellulose is exposed, a concentration of 9 per cent. is used. A concentration of 11 per cent. is required for examination of damage due to heat, light, chemicals or mildew, because this weakens the cuticle by splitting it, but without tearing it. Damage to the cuticle due to mild processing is slight, and therefore necessitates a concentration of 18 per cent., and this serves also for estimating the percentage of abnormally-thickened fibres. The investigations of Calvert and Summers (*J. Text. Inst.*, 1925, **16**, T. 233) on the mechanism of swelling by alkali are summarised, and a graph, reproduced from their paper, shows that as the concentration increases up to 13.5 per cent. the width of the fibre gradually increases, although with higher concentrations it remains constant on account of the constricting effect of the cuticle, the first influence of which is apparent at concentrations of 11 per cent. Similarly, stage (5) serves to swell the differentially-stained fibres and to accentuate colour contrasts. The examination should be made with a 2/3 inch objective and 10× eyepiece, with full daylight illumination. The mounts are only temporary, but will last a few days if sealed with a mixture of lanolin and colophony resin (1 : 4). The test is based on the different rates of diffusion of the dye into the exposed swollen secondary cellulose and cuticle of the fibre, with the result that the former is stained to a deeper shade; this diffusion depends on the nature and extent of the damage which the fibre has undergone, and is most critical at room temperature. The appearance of fibres which have been treated in various ways is described in detail and illustrated by photomicrographs. Mechanical damage may be evident from signs of surface bruising, deep cuts in the fibre wall, abrasion and broken fibre-ends. The test can also be used to indicate the nature and extent of the wear of finished goods, *e.g.* to distinguish between normal wear by frictional rubbing and contact with an abrasive surface. With heat tendering the cuticle breaks up, first into coarse spirals (*e.g.* after 24 hours at 95° C.), and then into finer spirals, until the constricting effect of the cuticle is no longer

Hemicelluloses of the Wood of English Oak. 5. Structure of Hemicellulose B.

M. H. O'Dwyer. (*Biochem. J.*, 1940, **34**, 149-152.)—The hemicellulose B from oak sapwood, like hemicellulose A from the same source, contains anhydroglucose units, whereas the hemicellulose B (and hemicellulose A) from oak heartwood contains none. The anhydroglucose units of sapwood hemicellulose B account for its positive specific rotation. All the hemicelluloses contain xylose and uronic acid residues, and on hydrolysis with takadiastase they yield xylose and a water-soluble

exerted and it becomes almost completely detached; the fibres are then stained uniformly bright red (e.g. after 19 days at 95° C.). Singed fibre-ends are shown by a brownish-red tip, unless chlorides are present, when the tips concerned do not take the stain. Acid tendering and over-bleaching result in changes in the cuticle similar to those produced by heat, but the spirals are less distinct, and transverse cracks appear in the final stages; bright red local blotches are characteristic of local tendering by strong acid. The test usefully supplements fluidity tests for the amount of bleached stock in a yarn, especially when the sample is small, and when a proportion of fully-bleached cotton in the loose state has been introduced in order to whiten a yarn. The results are roughly quantitative, and those obtained by different observers are in good agreement. If the presence of stock which is not fully-bleached is suspected, an ash-content determination is used to decide whether a wet treatment has been used, and the Congo red test serves to indicate its severity. Tendering effects due to light are similar to those caused by acid tendering, but are more localised, and mercerisation increases the depth of shade of the cuticle. Mildew effects are difficult to distinguish from mechanical and chemical damage, but there is more evidence of disintegration, and the hyphae, if present, are stained bright red; fluidity tests in cuprammonium solution should be used to confirm the conclusions. Bacterial tendering varies with the nature of the organisms; some produce very characteristic effects (e.g. fuzzy fibre-contours), whilst others behave similarly to mildew. "Dead" cotton, due to attack by fungus while the fibres are growing on the boll, is stained bright red in the Congo red test, and can thereby be differentiated from fungus attack of other (e.g. genetic or physiological) origin which produces a pink stain. In the examination of yarn breaks (*cf.* Clegg, *id.*, 1926, 17, T. 591) the number of broken fibres may be estimated by difference from the average numbers of fibres in the cross-section of the yarn, and of those which end naturally in the portion of the yarn in which the break occurs. A number of specific examples of applications of the test to the examination of defects in cotton goods, are described in detail and illustrated. J. G.

Use of Cheap Filter Paper for Analysing Tan Liquors. H. B. Merrill and G. C. Mueller. (*J. Amer. Leather Trades Chem.*, 1940, 35, 289-290.)—The official A.L.C.A. methods for the analysis of tanning extracts and yard liquors specify the use of either Munktell No. 1F or S. & S. No. 590 filter-papers. As these are expensive and difficult to obtain in war-time, the use of a cheaper grade of paper for routine analyses of yard liquors has been considered; in the present instance the substitute was a Central Scientific Co. No. 13250 paper, but it is suggested that many other cheap grades would be found equally suitable. Duplicate determinations of the soluble and

non-tannin matters and tannins in 14 representative yard liquors with the C.S. paper, as compared with the 1F paper, gave maximum differences of -0.06, -0.04 and -0.05 and average differences of ± 0.021 , 0.014 and 0.018 g. per 100 ml., respectively. These are regarded as negligible for routine purposes. Filtration with the cheaper paper was somewhat slower, although the yard liquors in question were relatively free from insoluble matter. J. G.

Inorganic

Titration of Nickel with Dimethylglyoxime with the Aid of the Dropping Mercury Electrode as Indicator Electrode. I. M. Kolthoff and A. Langer. (*J. Amer. Chem. Soc.*, 1940, 62, 211-218.)—The "amperometric" titration of nickel with dimethylglyoxime can yield accurate results and is applicable to dilute nickel solutions. Delicate electrical apparatus is required and the titration requires to be carried out in absence of air. Numerous other ions interfere. S. G. C.

Determination of Lead in Tin Coatings. P. Foerster. (*Ann. Chim. anal.*, 1940, 22, 93-94.)—The coating is dissolved off the specimen with hot water to which portions of sodium peroxide are added. The liquid is boiled to destroy peroxide, and the lead, together with small amounts of copper and iron, is precipitated as sulphide by the addition of sodium sulphide. The precipitate is filtered off, washed with dilute sodium sulphide solution, and dissolved in hydrochloric acid containing bromine; after removal of bromine by heating, the lead is re-precipitated as sulphide from alkaline solution in presence of potassium cyanide to keep copper and iron in solution. The lead sulphide precipitate is filtered off and dissolved, and the lead is determined by the chromate colorimetric method. S. G. C.

Improved Form of the Formaldoxime Colorimetric Method for Manganese. C. P. Sideris. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 307.)—The author's formaldoxime colorimetric method for the determination of manganese (*Ind. Eng. Chem., Anal. Ed.*, 1937, 9, 445; *Abst., ANALYST*, 1937, 62, 831) was found inadequate by Wiese and Johnson (*J. Biol. Chem.*, 1939, 127, 203; *Abst., ANALYST*, 1939, 64, 459), and, in more recent studies by the author, for biological material of high phosphate-content. The interference of phosphates can be eliminated by precipitation with lead acetate in acetic acid solutions, and the original method may then be employed. An aliquot portion (10 ml.) of the solution of ashed biological material in dilute hydrochloric acid is neutralised with *N* sodium hydroxide solution, the amount of alkali required being ascertained by titration of another aliquot portion with methyl red as indicator. The solution is then acidified with 2 ml. of 20 per cent. acetic acid, which dissolves the manganic phosphate but not the ferric phosphate precipitate. Excessive phosphate, e.g. with plant

tissues, is removed by the addition of 0.5 ml. of a 5 per cent. solution of lead acetate. The mixture is agitated and allowed to stand for 10 minutes, and excess of lead salt is removed by the addition of 1 ml. of a 20 per cent. sodium sulphate solution. After 30 minutes the mixture is filtered or centrifuged. An aliquot portion (10 ml.) of the filtrate is neutralised with 40 per cent. sodium hydroxide solution, 3 to 4 drops of the formaldoxime reagent (*Sideris, loc. cit.*) are added and then more of the 40 per cent. sodium hydroxide solution until the colour develops. The addition of sodium cyanide, as prescribed in the original publication (*loc. cit.*), is not necessary. Ferric chloride need not be added to the comparison standard solution if the removal of iron from the sample solution was complete. The final volume of the sample and comparison solutions is made up to 15 or 20 ml. In 10-ml. portions of sample containing from 0.005 to 0.01 mg. of manganese in presence of 0.010 to 0.100 mg. of phosphate (PO_4) the average error was 2 per cent. A. O. J.

Volumetric Determination of Small Quantities of Barium and Sulphate with Barium Rhodizonate as Indicator. C. C. Miller. (*J. Chem. Soc.*, 1940, 401-406.)—

Advantages are offered by the use of a suspension of the scarlet modification of barium rhodizonate as indicator; 40 mg. of sodium rhodizonate are dissolved in 20 ml. of water and treated with 0.1 g. of barium chloride dissolved in a little water. The flocculent brown precipitate is centrifuged, washed with a little water and then with ethyl alcohol; 5 ml. of ethyl alcohol, containing 0.1 ml. of conc. hydrochloric acid, are added to the precipitate, and the tube is placed in hot water to convert the complex into the scarlet form. The precipitate is centrifuged off and suspended in 250 ml. of alcohol. The suspension keeps well in a stoppered bottle; it is shaken before use.

Titration of Barium.—The solution (10 ml., equivalent to 2 to 20 mg. of sulphate and containing 0.2 to 1 ml. of 2 N hydrochloric acid) is treated with 0.5 ml. of indicator suspension, and alcohol equivalent to the volume of the solution, less 5 ml., is added. The mechanically stirred liquid is titrated, drop by drop, with a 0.02 M solution of ammonium sulphate in 50 per cent. alcohol until the pink colour disappears. A very slow rate of titration (1 drop in 10 seconds) is advisable near the end-point, and the temperature is preferably 25° C. Occasionally a pale pink colour persists beyond the end-point, owing to adsorption on the barium sulphate. The most consistent results were obtained with solutions containing 0.5 ml. of 2 N hydrochloric acid in a final volume of 18 to 27 ml. **Determination of Sulphate.**—Direct titration of sulphate is impracticable. The following indirect method was devised:—Ten ml. of the sulphate solution, containing 2 to 20 mg. of sulphate and 0.5 to 1 ml. of 2 N hydrochloric acid, are titrated at 90° C., at the rate of 1 ml. per minute, with 0.02 M barium chloride solution

until an excess of 1 to 2 ml. is present. The precipitate is digested at 90° C. for 10 minutes, and the liquid is then cooled. Alcohol equivalent to the total volume, less 5 ml., is added, followed by 0.5 ml. of indicator suspension, and the liquid is titrated with 0.02 M ammonium sulphate solution in 50 per cent. alcohol, as in titrating barium. **Notes.**—The required degree of acidity for the titration was conveniently obtained by the use of "brilliant cresyl blue" indicator. The indicator solution, as supplied by the British Drug Houses, was diluted tenfold with alcohol, and 0.1 ml. of the dilute solution was added to the solution to be titrated. Sufficient 2 N hydrochloric acid was added until the blue colour was just discharged; this corresponds to the addition of about 1 ml. of the acid to a neutral solution. For solutions initially too acid, the acidity was conveniently reduced by addition of magnesium acetate solution until the blue colour appeared and then just discharging it with 2 N hydrochloric acid. The addition of alcohol causes the blue colour to return. In association with the scarlet barium rhodizonate the blue produces a violet colour changing at the equivalence point to a pure blue. The change-point was sharper than in absence of the blue indicator. The accuracy of the determination was within about 1 per cent. of the theoretical amount. The only ions found to have any appreciable interfering action were ferric, cupric, lead, calcium and phosphate. Reduction with hydroxylamine hydrochloride or aluminium powder overcame the interference of ferric and copper salts. Sodium rhodizonate costs 5s. per g., but 40 mg. converted into barium rhodizonate suspension suffices for 500 titrations. S. G. C.

Removal of Phosphoric Acid from Hydrogen Peroxide. S. R. Dickman and R. H. Bray. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 279.)—Phosphoric acid present in hydrogen peroxide as a stabiliser, may be removed, if objectionable, by the following process:—Ten ml. of 2 per cent. ferric chloride solution and 5 g. of calcium carbonate are added to a 100-ml. sample, and the mixture is stirred and filtered on a Buchner funnel. The hydrogen peroxide solution is then acidified with an alternative acid for stabilisation. S. G. C.

Microchemical

Colorimetric Determination of Silver. C. G. Makris and R. Menaché. (*Ann. Chim. anal.*, 1940, 22, 117-120.)—The method is based on reduction with tannin in presence of small amounts of alkalis and a solution of sodium albuminate. The test substance should first be converted into nitrate and then dissolved in 2 ml. of water in a small colorimeter tube. Three different amounts (0.25, 0.5 and 1 ml.) of a standard 0.01 per cent. silver nitrate solution are measured into other tubes, and each is made up to 2 ml. Into each tube are introduced 4 ml. of a freshly prepared 0.5 per

cent. solution of tannin and 0.18 ml. of 0.1 *N* sodium hydroxide solution, and then, after shaking, 2 drops of sodium albuminate (5 ml. of fresh egg-white in 100 ml. of 0.1 *N* sodium hydroxide solution). After 5 minutes the tubes are examined, the unknown being compared with the standard solution most approximating to its colour intensity. Lead, if present in large proportion, should be removed as sulphate before determining the silver. The average error is ± 5 per cent. J. W. M.

Colorimetric Determination of Magnesium. C. P. Sideris. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 232-233.)—The method is devised for the determination of magnesium in dried plant tissues. The material (2 g.) is ashed in a platinum or porcelain crucible, and the ash is dissolved in 5 ml. of 18.5 per cent. warm hydrochloric acid. After dilution to a suitable volume (100 ml.) any insoluble matter is removed by filtering or centrifuging. An aliquot part is transferred to a 50-ml. centrifuge tube, treated with 1 ml. of 1 per cent. lead acetate solution to precipitate phosphate, and centrifuged. The excess of lead is precipitated with 1 ml. of 1 per cent. sulphuric acid and separated by centrifuging. The filtrate is transferred to a 125-ml. conical flask and neutralised with *N* sodium hydroxide solution, methyl red being used as indicator. The calcium is precipitated with 2 ml. of 20 per cent. acetic acid and 5 ml. of 4 per cent. ammonium oxalate. After standing 4 to 6 hours the mixture is filtered through asbestos, and the residue is washed with water. If required, the calcium may be determined, after solution, by titration with permanganate. To the filtrate are added 2 ml. of 20 per cent. acetic acid and 3 ml. of a 5 per cent. alcoholic solution of 8-hydroxyquinoline. If a precipitate forms (copper or nickel hydroxyquinolate) the mixture is centrifuged and the precipitate is filtered off and discarded. To the clear solution 5 ml. of conc. ammonium hydroxide (28-29 per cent.) are added, and the mixture is heated over a water-bath for 30 minutes. It is then cooled and treated with about 5 ml. of chloroform to dissolve the hydroxyquinolates of iron, manganese, aluminium, etc. After an hour the mixture is filtered through a G4 Jena sintered glass filter, which is rinsed with at least 5 ml. of a 10 per cent. aqueous solution of ammonium hydroxide and 2 ml. of chloroform. The magnesium hydroxyquinolate is dissolved in 5 ml. of 18 per cent. hydrochloric acid and made up to a known volume. An aliquot amount (10 ml.) is buffered with 3 ml. of a 50 per cent. solution of sodium acetate, and 3 ml. of a 1 per cent. solution of ferric chloride hexahydrate are added. The green-black pigment is extracted with chloroform (3-4 times), made up to volume with butyl alcohol (to retard volatilisation), and compared in a colorimeter with a standard solution, prepared in the same way, containing 10 to 20 γ per ml. Results were excellent with amounts of magnesium ranging from 1 to 0.1 mg. J. W. M.

Test for Iodide and Nitrite. C. L. Wilson. (*Chem. and Ind.*, 1940, 59, 378-379.)—Several brands of special paper sold for spot tests contain enough starch to enable the usual iodide test to be carried out without any specially-prepared starch solution. In making the test a drop of 2*N* acetic acid is placed on the paper, and the test drop is run in from a capillary and followed by a drop of 0.1*N* potassium nitrite solution for the oxidation. *Limit of identification:* 0.025 γ of iodide; *concentration limit:* 1 : 2,000,000. In absence of other oxidising agents the test may be used reciprocally for nitrite. *Limit of identification:* 0.005 γ of NO₂; *concentration limit:* 1 : 10,000,000. J. W. M.

Microgram and Millimicrogram. (*Ind. Eng. Chem., News Ed.*, 1940, 18, 491.)—The Committee of the American Chemical Society on Nomenclature, Spelling, and Pronunciation has approved the following recommendation of the Division of Microchemistry:—For 0.001 milligram the term "microgram," designated by the symbol γ (the word "gamma" should not be used as a substitute for microgram). For 0.001 microgram, the term "millimicrogram," designated by the symbol $m\gamma$ (the term "milligram" is not to be used).

Determination of Sodium in Biological Fluids. M. C. Darnell, Jr., and B. S. Walker. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 242-244.)—Either 0.1 ml. of material wet-ashed by means of nitric and sulphuric acids and hydrogen peroxide, or an equivalent amount of the trichloroacetic acid filtrate are analysed, the total volume by either method being 1 ml.; this is placed in a 15-ml. conical centrifuge tube. To the sample solution 5 ml. of freshly filtered zinc uranyl acetate reagent are added (for preparation, see Wembach, *J. Biol. Chem.*, 1935, 110, 95) and then at 5-minute intervals seven 0.3-ml. portions of 95 per cent. ethyl alcohol, with careful tapping of the bottom and thorough rotatory mixing. The last two additions are allowed to remain on top, and the tube is centrifuged at 2000 r.p.m. for 10 minutes, and, after decantation of the liquid, is inverted, allowed to drain, and treated with 2 ml. of wash liquid (300 ml. of ethyl acetate of analytical reagent quality, diluted to 1 litre with glacial acetic acid); this process is repeated. Finally, 5 ml. of ether are used for washing, 5 minutes being allowed for centrifuging and 1 minute for draining. **Blood and cerebrospinal fluid.**—The dried precipitate is dissolved in 4 to 5 ml. of water, and the solution is transferred to a 100-ml. flask. It is then diluted to about 70 ml., and 4 ml. of 5 per cent. sulphosalicylic acid, 4 ml. of a 10 per cent. solution of sodium acetate trihydrate, and water to make 100 ml. are added. The colour is determined photometrically by means of an Evelyn colorimeter tube and a 440 $m\mu$ filter. The instrument is set to read 100 with a tube containing 4 ml. each of 5 per cent. sulphosalicylic acid solution and 10 per cent. sodium

acetate solution in 100 ml. The sodium-content of a blank similarly treated must be subtracted. *Urine*.—The precipitate is dissolved in 10 ml. of water. Any uranyl phosphate present remains in the form of a gelatinous precipitate. The tube is centrifuged for 10 minutes at 2000 r.p.m., and 5 ml. of the

supernatant liquid are transferred to a 50-ml. graduated flask, diluted to 35 ml., and treated with half the quantities of the reagents used for blood and cerebrospinal fluid, and the readings are taken as before. The maximum error in the authors' determinations did not exceed 1 per cent. J. W. M.

Reviews

MELLOR'S MODERN INORGANIC CHEMISTRY. 9th Edition. Revised and edited by G. D. PARKES, M.A., D.Phil., in collaboration with J. W. MELLOR, D.Sc., F.R.S. Pp. xix + 915. Longmans, Green & Co., Ltd. 1939. Price 12s. 6d.

On reading this book one is immediately impressed with two things. The first is its surprisingly low price, when the ground it covers is taken into consideration; the second is the profound yet delicate historical sense of its authors. This may seem a strange quality to select first in a criticism of a scientific textbook, but in point of fact it has done more than provide a brief historical introduction, a good deal of interesting and out-of-the-way information concerning the many minerals mentioned, and arresting quotations, drawn from many sources and ages, at the headings of the chapters; it has made of the whole of the first section of the book a veritable *tour de force*. This first section (of 250 pp.) deals with the theories of chemistry and, maintaining the historical sequence, presents them in a logical and clear manner which is masterly; where the facts on which theories are based seem to make possible some other explanation, this is stated, concrete examples of actual weights, volumes and so forth are given, and the reader finds himself in touch with the authors in a somewhat unusual manner. The next 287 pages deal with the elements hydrogen, oxygen, carbon, nitrogen, sulphur and the halogens and then follow the remaining elements in their periodic groups, including excellent discussions of family relationships at the end of each. In this descriptive part the high inspiration of the earlier chapters seems to "fade" somewhat "into the light of common day" of the ordinary textbook—perhaps this is inevitable, and in any event the standard remains good. Possibly the defects of the historical sense are here making themselves felt, and the criticism is one, indeed, that applies to a very large proportion of textbooks; but one cannot help wondering whether the time is not ripe for the consignment of any detailed discussion of, for example, the hardness of water (with its pendant jewel, Clark's soap test) to more specialised textbooks. Whether or no this is so, the proper place for the discussion of hardness is surely not under water *considered as a compound of hydrogen*. Other changes in traditional presentment also suggest themselves; is it not also time that details of ore smelting and like processes, which are primarily the concern of metallurgical students, should largely be left to them, and, in any event, why give processes which are outworn when those processes are of no special chemical interest? The fault lies probably not with these or any other authors, but with examiners, for whose demands they have to cater and who do not seem to realise that inorganic chemistry has grown to huge proportions and that the padding of an earlier day should now be stripped from it. The times are changing, and many an element which was a "chemical curiosity" a few decades ago has become an important factor in the life of nations and so demands far more than the few lines of small type that is all that subjects such as hardness of water, the Leblanc process and various methods of steel-making now allow for it. As so often happens with textbooks, among the least satisfactory portions are those devoted to analytical chemistry; they are short and make no pretence to be exhaustive and so little harm

is done, but it is not provocative of confidence to be told (p. 796) that *heating* a solution of steel in nitric acid with sodium bismuthate oxidises all the manganese to permanganate—this being the one thing you must not do with it. One curious omission must be recorded: the pardonable enthusiasm of the authors in describing the discovery and separation of the rare earths has led them to forget to give any description of cerium at all.

At the end of the Group VIII elements there is an account of Werner's valency theory, which reverts to the clarity of the beginning of the book; the same may also be said of the account of radio-activity in the final chapter. The book ends with a series of questions drawn from English-speaking universities of many lands, and (an unusual feature) answers to these where they may be given briefly. The volume is well produced and bound, but is not entirely free from misprints, proof-reading slips and misuse of words; these, however, are relatively unimportant. Such criticism as has been made is very slight in comparison with the outstanding merit of the book; its eight predecessors have shown by their unexampled success how adequately they filled a gap, and one can pay no higher tribute to this, the ninth edition, than to express the sincere belief that it will carry that success to still higher peaks. One can but regret that one of the authors has not lived to see it.

B. S. EVANS

PHOTOGRAPHY BY INFRA-RED: ITS PRINCIPLES AND APPLICATIONS. By W. CLARK, Ph.D., F.I.C. Pp. xi + 389. London: Chapman & Hall. 1940. Price 25s. net.

There is an extensive and ever-growing literature on ultra-violet rays and their applications, but, with the exception of a few small practical handbooks, there has hitherto been no complete textbook on the rays just beyond the other end of the visible spectrum. Although nearly 140 years have passed since Herschel discovered infra-red rays, the difficulty of studying them stood in the way of their general scientific application. Even after Higgs, in 1891, had prepared an emulsion sensitive to the extreme red, photography by infra-red rays remained in the experimental stage. It was not until the discovery of the use of dicyanine as a sensitiser (since replaced by kryptocyanine and neocyanine) that it became practicable to apply the method in special investigations. Since 1934 the introduction of tetra- and penta-carbocyanines as sensitisers has made it possible to produce plates sensitive up to 13,000 Å, but for most practical purposes a sensitivity between 9500 and 10,800 Å has proved sufficient.

In this book theory and practice go hand in hand. Dr. Clark first gives an outline of the theory and development of infra-red photography, including a valuable description of the evolution of sensitisers, with practical directions for sensitising plates. He next discusses the Herschel effect and indirect methods of infra-red photography, and then (Chapter VII) gives a full and practical account of the sources of infra-red radiation. These general chapters are followed by detailed descriptions of methods for the examination and differentiation of materials by means of infra-red rays. Among the subjects dealt with and illustrated in this chapter are the examination of textiles, paintings, inks and pigments, and documentary photography, including the deciphering of charred papers and erased writing.

Chapter IX gives a full description of the application of infra-red photography to medicine, and includes the study of the superficial venous system and the transmission and reflection of the rays by skin and tissue. Chapter X deals with botany and palaeontology, and Chapter XI with infra-red photomicrography. Chapter XII, which describes special applications of infra-red photography, is perhaps the most striking from the popular point of view; it includes general landscape photography, photographic survey from land and air, fog penetration, and the photography of hot objects. The last three chapters have for their subjects

the measurement of infra-red radiation, the penetration of radiation through the atmosphere, and the optical characteristics of materials.

After each chapter there is a full bibliography containing, in all, some hundreds of references, and the book concludes with good name- and subject-indexes. The numerous illustrations are excellent, and the printing is clear and easy to read.

It would be difficult to overpraise this book. It is much more than a compilation of the work of others, for it is imbued throughout with the author's own knowledge and experience. It can be warmly recommended as a standard textbook on a fascinating subject about which there is still much to learn. EDITOR

STARCH AND ITS DERIVATIVES. By J. A. RADLEY, M.Sc., A.I.C. Pp. 346, with 61 photomicrographs and 28 figures. London: Chapman & Hall, Ltd. 1940. Price 22s. net.

This volume is the eleventh in the series of monographs on applied chemistry, produced under the editorship of Dr. E. Howard Tripp. Its contents have been divided into four parts. The first of these is a very interesting one dealing with the structure of the starch molecule, the physical chemistry of starch, the reaction of starch with iodine, and starch esters. A survey of the history of starch and dextrin is included, from which it is amusing to learn from well-authenticated reports that the discovery of dextrin was made by a method similar to that which led to the Chinese discovery of roast pork. Particular attention has been devoted to the physical properties of starch, because these have considerable importance in industry. Part II deals with the manufacture of various root and cereal starches, with modified starches, and with some starch derivatives—glucose, maltose, ethyl alcohol and dextrin. Part III is of more particular interest to the manufacturer. The field covered is large; in addition to an account of adhesives made from starch and dextrin, the uses of starch in the paper and textile industries are discussed and a useful section on enzymes used in the starch industry is included. Part IV is the one that should be of most use to the analytical chemist. It opens with a discussion on the general examination of starches, deals with the determination of starch and the analysis of dextrin, and concludes with methods of determining the activity of enzyme preparations.

Particular praise is due for the admirable series of photomicrographs, all but a few of which are in duplicate, showing the same field under ordinary and polarised light. The reproduction of these is as good as may be found anywhere and reflects much credit on both printer and publisher. The proof-reading also has been excellent, for there are very few errors and those which have been detected are innocuous.

The book concludes with a subject index and a name index, both of which seem adequate. In the latter there is some confusion between brothers, the work of H. R. Nanji being indexed under the name of D. R. Nanji, though in the references in the body of the book both names are correctly coupled with the two authors' contributions to the literature of starch, except in two places where, in mention of the work of J. J. Chinoy, F. W. Edwards and H. R. Nanji, the last name is omitted.

The value of this book to chemists, research workers and manufacturers lies mainly in its wealth of references. No important contribution to science appears to have been omitted, and they constitute an invaluable guide not only to those parts of the subject already adequately covered but to those in which there is much need for further research.

This volume, like its companions in the series, is much more than one of the "Recent Progress" order, but even if it is viewed merely in that light, it can be warmly commended to all interested in the chemistry of starch and allied substances.

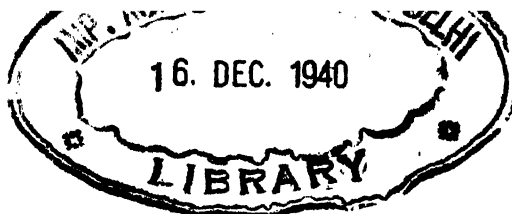
F. W. EDWARDS

BREWING SCIENCE AND PRACTICE. VOL. II., BREWING PROCESSES. By H. LLOYD HIND, B.Sc., F.I.C. Pp. xiv + 514 with 157 illustrations. London: Chapman and Hall. 1940. Price 56s.

The first volume of this comprehensive work was reviewed in *THE ANALYST* (1939, 64, 149), and the second volume has now appeared after some delay due to war conditions. The original intention of the author was that the book should be completed in two volumes, but the inclusion of the results of recent investigations of considerable importance has necessitated the production of a third, which will deal with bottling, cleaning, brewery by-products and analysis.

The contents of the present treatise comprise descriptions of the construction and care of the many varieties of plant used throughout the brewing process from the grinding of the malt to the racking of the finished beer into casks. Each stage of the process, together with the selection of the water, malt, hops and yeast used for the production of different types of beer, is dealt with, and the effects upon the composition of the worts and beers of modifying the various conditions are discussed in considerable detail. The more scientific sections of the work include the biochemical or physical changes occurring during mashing, boiling, cooling and fermentation, the microbiology of deleterious organisms such as "wild" yeasts, moulds and bacteria (including an excellent chapter written by Dr. J. L. Shimwell on beer bacteria, based largely upon his own valuable researches during recent years) the nutrition and respiration of yeast, the redox potential of beer and its determination, and finally the causes of non-biological turbidities of beer. The numerous illustrations add considerable value to the volume, for whether, as in the form of plates, they depict photographs of brewery plant or photomicrographs by visible and ultraviolet light of yeast or other organisms, or appear as line blocks in the text, they are well selected and admirably reproduced, with perhaps one exception, Fig. 220, in which loss of detail has eliminated many spectrum lines. The experience of the author, both as brewer and chemist, has enabled him to summarise the whole available knowledge in an extensive field of industry with wide ramifications in detail, and the present volume shows the many admirable qualities of its predecessor. Numerous results of scientific research, which are here published in book form for the first time, will be welcomed by brewing chemists and others interested in the physical, chemical and biochemical sciences underlying a great industry.

To the brewer this volume will naturally appeal to a greater extent than the previous one, and its sound and comprehensive character will ensure it a position in the brewing room library, where it will serve as a valuable and up-to-date book of reference on all matters pertaining to the art of beer production. T. J. WARD



THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

Obituary

JOCELYN FIELD THORPE

JOCELYN FIELD THORPE was born in Clapham, London, on December 1st, 1872, the sixth son of William George Thorpe, F.S.A., of the Middle Temple. He received his early education at Worthing College, and in 1888 entered King's College, London, as an engineering student under Professor Wilson. The two years spent in the engineering laboratories, and particularly the experience derived from the shops at Alexander Wilson's Vauxhall Works during each long vacation, appealed to Thorpe's practical sense and laid a foundation of experience in engineering science which he found of great service in his future career as a chemist. His connection with King's College was appropriately recognised when he was elected a Fellow of the College in 1924.

It was a somewhat revolutionary step in those days to choose chemistry as a profession, but this bold course was decided upon by Thorpe's father on the advice of Sir Edward Thorpe, in whose department, at the Royal College of Science, South Kensington, he then entered as a student in 1890. Here he remained for two years, taking only the first year course of elementary chemistry and physics and the third year of advanced chemistry, chiefly organic chemistry, in which he found his vocation. In 1892, again on the advice of Sir Edward Thorpe, he entered the research school of Victor Meyer at Heidelberg, at that time the leading school in organic chemistry; he worked there for two years under the direction of Carl Auwers, and obtained the degree of Ph.D. in 1895.

On his return to England, Thorpe went to Owens College, Manchester, where he remained for ten years. At first he was a Research Fellow and obtained the degree of D.Sc.; later he became a member of the teaching staff. He carried out much important work at this period, a good deal of it in connection with the synthesis of degradation products of the terpenes, which was attracting considerable attention at that time. Towards the end of this period in Manchester, Thorpe was elected a Fellow of the Royal Society. In 1902 he married Lilian, the only daughter of the late William Briggs, J.P., of Hale, Cheshire.

In 1909 Thorpe was awarded the Sorby Research Fellowship of the Royal Society, and spent four years at the University of Sheffield, working mainly on various problems of aliphatic chemistry, such as the isomerism of the glutamic acids and the reactions of the imino-compounds, including the reaction which bears his name.

In 1914 he was appointed Professor of Organic Chemistry, University of London (Imperial College), a post he occupied until his retirement in 1938. As soon as he was appointed he set out to expand the then limited facilities for research; this expansion was necessarily interrupted by the war, but he came back to the task in 1919 and founded a school of research second to none in the

country. His enthusiasm and encouragement stimulated the activity of a large band of pupils and attracted research students not only from home and the Dominions but also from foreign countries.

Thorpe's ability and shrewd common sense were well known, and many public bodies were glad to avail themselves of his services. Thus he was member of the Chemical Defence Committee, War Office, and in 1917 received the honour of C.B.E.; he served on the Advisory Council, Department of Scientific and Industrial Research, 1916-22. He was President of the Indian Chemical Services Committee, 1919-20, member of the Safety in Mines Research Board, 1924-35, and Chairman of the Explosives in Mines Committee, Department of Mines; and member of the Dystuffs Development Committee, Board of Trade, 1925-34.

Thorpe gave many years of devoted service to the Chemical Society, of which he was in turn Treasurer, Vice-President and President during the important period of 1928-31.

He was also a Member of Council of the Royal Society 1923-25, and President of the Institute of Chemistry 1933-36. He was awarded the Longstaff Medal of the Chemical Society in 1921, and the Davy Medal of the Royal Society in 1922. In 1933 he was elected an Honorary Member of this Society. In 1939 a Knighthood was conferred upon him in recognition of his valuable public services.

Thorpe owed much of his success, both as a teacher and as a man of affairs, to his personal qualities, his joviality and charm of manner. He had an incorrigible faith in the goodness of human nature and refused to see anything but the best in the people with whom he came in contact. His kindly interest in his old students will also be widely remembered. He was an admirable host, in which capacity he was ably supported by Lady Thorpe, and a large circle of friends will long cherish happy memories of their kind and gracious hospitality.

On his retirement, as Professor Emeritus, from the Imperial College, Thorpe found great happiness in his delightful home at Cooden Beach; here he carried on his literary work and the duties connected with numerous committees of which he was an active member. He had a great love for, and a wide knowledge of, old English china, of which he made a valuable and unique collection; he also took a keen interest in his garden, especially in the growing of roses. The end came peacefully, but with tragic suddenness, on June 10th, 1940, a few hours after he had devoted the usual daily attention to his rose garden.

M. A. WHITELEY
G. A. R. KON

Spectrophotometric Assay of Vitamin A, with Special Reference to Margarine

By J. R. EDISBURY, B.Sc., PH.D.

(Read at the Meeting, May 1, 1940)

THE recent Regulation of the Minister of Food, requiring all margarine sold in the British Isles to be vitaminised, will have directed attention to the problems involved in the assaying of vitamin concentrates sold for this purpose and of the vitaminised margarines offered to the public.

As with all physiologically active materials, the biological approach to the problem of assaying is an indispensable first step and the ultimate court of appeal in cases of doubt, but in ordinary practice it is open to objection on account of the time required for an assay, the highly specialised technique involved and the relatively large experimental error inseparable from biological methods. Those concerned with the assaying of vitamin A preparations are fortunate in having available chemical and physical means for securing the desired information, and

in being able to link up such observations with the biological effect of the vitamin. None of these methods is free from difficulty, and where the degree of dilution is high—as in margarines and butters—special skill and experience are needed both in carrying out the tests and in interpreting the results.

The physical assay of vitamin-A concentrates may be undertaken by any, or all, of the following well recognised means:—(1) Blue value in various guises (Carr-Price,¹ Moore Blue Units,² Dilution Strength,³ Dann and Evelyn L-620 $m\mu$ value⁴). (2) Vitameter⁵ and simple photoelectric devices (*e.g.* McFarlan *et al.*⁶). (3) Spectrophotometric technique.

The first of these depends on measurement of the intensity of blue colour developed on adding a solution of antimony trichloride in chloroform to a solution of the concentrate, also in chloroform. The second depends essentially on measuring the intensity of light absorption in the ultra-violet at a wavelength of 325 $m\mu$, the vitamin A absorption maximum. The spectrophotometric method, essentially a combination and refinement of the first two methods, is of special value in that the tests are complementary and provide additional evidence of normality or abnormality—a matter of importance in the interpretation of results. From a spectroscopic point of view, the “blue value” determination involves measurement of the intensity of light absorption at 620 $m\mu$, and to support this observation, measurements at 580 $m\mu$ are also made. Margarines in particular respond better to spectrophotometric treatment than to other methods, although with careful attention to detail (including “calibration” of the observer) an abridged form of the “dilution test” described by Andersen and Nightingale³ is specially useful in routine checking.

It is proposed here to describe briefly the application of spectrophotometric methods to the assay of vitamin A in margarines and the “concentrates” used in their vitaminisation.

APPARATUS.—For a comprehensive assay, two instruments are required, *viz.* for the colour test, a visual spectrometer with photometer; for the ultra-violet examination, either—and preferably—a medium quartz spectrograph and photometer, taking 10 in. \times 4 in. plates, or a vitameter. Provided that the limitations inherent in measurements confined to 325–330 $m\mu$ are recognised, and that the instrument is adapted for photography⁷ and calibrated spectrometrically, the Hilger vitameter affords an inexpensive alternative to the ultra-violet spectrophotometer, and is very nearly as accurate in use (probably ± 3 per cent.).

SOLVENTS AND REAGENT.—Special attention should be given to the choice of solvent for dissolving the material under ultra-violet examination, and to the preparation of antimony trichloride reagent. The solvents normally in use for ultra-violet work are cyclohexane, ethyl and *isopropyl* alcohols, etc., and between these solvents the choice usually depends almost entirely on convenience. Chloroform or ether should be avoided.^{8,9} For general purposes cyclohexane may be safely recommended; *isopropyl* alcohol has very occasionally been found to give reproducible results a few per cent. higher than cyclohexane. For measuring the absorption at 620 $m\mu$, which is characteristic for normal vitamin A when treated in solution with antimony trichloride, it is essential to use cyclohexane or chloroform as solvent—the alcohols obviously cannot be used.

We have found that special care is necessary in the preparation of the antimony trichloride reagent, to ensure removal of any trace of ferric chloride. The standard 20–25 per cent. solution of antimony trichloride in chloroform should be purified by adding 0.1 per cent. of water, shaking thoroughly and leaving in a refrigerator overnight before decanting the clear reagent; this should be kept in a colourless glass bottle of good quality, painted on the outside to exclude light. Coloured glass bottles give up sufficient iron to spoil the reagent.

EXAMINATION OF CONCENTRATES.—The general procedure is varied according to the character of the material. For concentrates, in which the potency may be

of the order of 1000–200,000 I.U. or more of vitamin A per gram, the recommended procedure is as follows:

Prepare a solution in pure cyclohexane and dilute as required; 1 per cent. (w/v) is suitable for a concentrate of 5000 I.U. per g., and concentrations in inverse proportions for materials of other potencies.

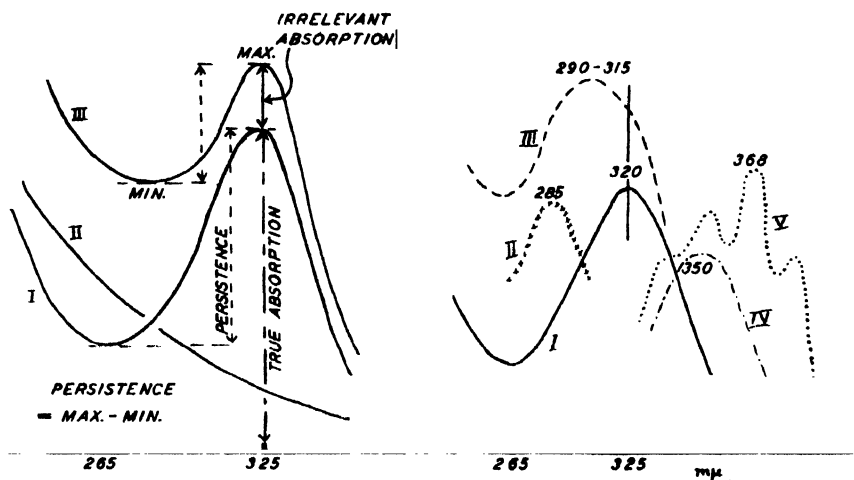
1. For *ultra-violet assay*, photograph the absorption spectrum of duplicate portions in two stratum thicknesses* that differ by about 10 per cent. (say 3 and 3.3 mm.) and determine the $E_{1\text{cm}}^{1\%}$ $325m\mu$ value. This represents the total light absorption at $325m\mu$ due to the combined effects of the vitamin A and the diluent oil, if any (Fig. 1). The absorption of the refined and

Fig. 1

IRRELEVANT ULTRA-VIOLET ABSORPTION

(a) Removed by saponification.

(b) Not removed by saponification.



- I. Vitamin A.
II. Glycerides, etc.
III. Observed summation curve.
Note diminished persistence.

- I. Vitamin A.
II. Unknown component of whale-liver oil (A_3 ?).
III. Typical commercial whale-liver oil = summation of I and II.
IV. Vitamin A_3 .
V. Cyclised vitamin A (crude).

deodorised oil often used varies from sample to sample over a range of about 0.07 to 0.2 at this wavelength, and must be deducted in order to obtain the true (net) vitamin contribution. If, as is most likely, a sample of diluent oil is not available for direct test† a standard deduction of 0.1 from the gross or total $E_{1\text{cm}}^{1\%}$ $325m\mu$ value will thus bring the result to within 0.1 of the true figure. Such correction is not required for values exceeding $E_{1\text{cm}}^{1\%}$ $325m\mu = 10$. With further dilution (by 5, in the example cited) the solution will be suitable for vitameter readings.

In the routine testing of a normal concentrate, more than this is not strictly required. The colour reaction forms a useful check, however, particularly when the result is doubtful, and in the examination of an unknown material it provides a convenient first approach from which appropriate conditions for ultra-violet assay can be calculated, or further treatment planned.

* Alternatively, one portion can be diluted slightly—say 10 per cent.—using an N.P.L. cylinder, and the same thickness employed again.

† If available, a 1–1.5 mm. thickness of the diluent oil is convenient, the concentration being taken as 92 per cent. for purposes of calculation.

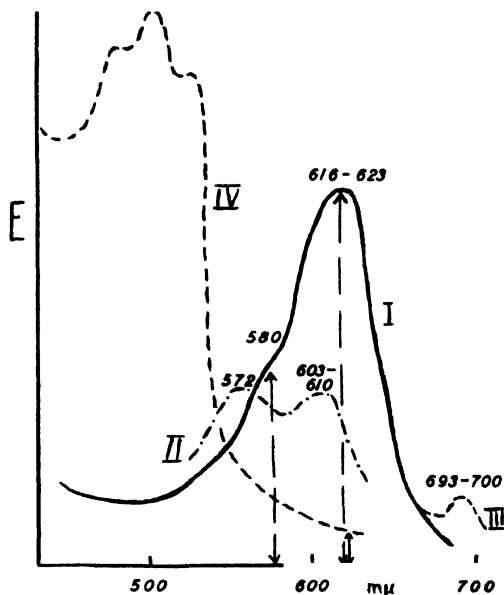
2. *Colour Test*.—The same cyclohexane solution can be employed. The presence of up to 15 per cent of cyclohexane has not been found to have any discernible effect on the antimony trichloride reaction, and the saving of time by the use of one solution is considerable.

The Carr-Price blue value is estimated in the standard manner, but one point requires emphasis: the solution must be diluted (N.P.L. cylinder) until under the accepted conditions of 0.2 ml. of solution and 2 ml. of reagent in a 1-cm. cell the colour *matches 5 Blue* on the Lovibond scale (plus the usual corrections, say $+1.5Y - 0.3N$). A low "match" gives high results, and *vice versa*.

In the spectrophotometric colour test, the cyclohexane solution (1 per cent. for 5000 I.U. per g.) is again used, but otherwise the technique devised by Morton⁸ is followed in essentials. Intensity readings are taken at the absorption maximum, 616–623 $m\mu$, and also at 580 $m\mu$, where

Fig. 2

ABSORPTION SPECTRUM OF ANTIMONY TRICHLORIDE COLOUR REACTION



- I. Normal vitamin A. Note 2 : 1 ratio between intensities at 620 and 580 $m\mu$.
- II. Severely inhibited vitamin A as in cod-liver oil. The ratio is altered and the wavelength-maxima displaced. On saponification, normality is restored. II and I are typical of the same weight of vitamin A before and after saponification of a weak oil.
- III. Vitamin A₂.
- IV. Complex formed between antimony trichloride and colouring matter of a typical margarine. This gives a relatively stable red colour, the 620 $m\mu$ absorption of which can be determined after the vitamin A band has faded.

in normal circumstances the intensity is about half that at 620 $m\mu$ (Fig. 2). Any marked deviation from a wavelength maximum of 620 $m\mu$ (say to 603–606 $m\mu$, as seen in the direct testing of cod-liver and similar oils) or from a bright blue colour, calls for further investigation. Reliable estimates of vitamin content cannot as a rule be based on uncorroborated data obtained under these conditions.

With a 2-cm. cell, matching at between 1.8 and 2.2 on the colour density ($\log I_0/I$) scale is recommended. As in the Carr-Price test, unreliable results are otherwise likely to be obtained. Cloudiness of the solution can usually be cleared by addition of a drop of acetic anhydride.

Even with all precautions, the absorption intensity or colour density at 620 $m\mu$ is often lower than might be anticipated from the ultra-violet results, probably owing largely, but not exclusively, to inhibition¹¹ (mechanical as well as chemical) of colour development by components of the diluent oil. Some typical results are summarised in Table I.

TABLE I
TYPICAL ULTRA-VIOLET AND COLOUR-TEST DATA

| | | | | $E_{1\text{cm}}^{1\%}$ | | | | | |
|--|--|--|--|------------------------|--------------|---------------------------|--------------|---------------------------|--|
| | | | | 620m μ | | Blue value | | | |
| | | | | 325m μ obsd. | obsd. | calcd. from obsd. u.v. | obsd. | calcd. from obsd. u.v. | |
| Normal rich fish oils or concentrates... | | | | { 50 32 | 155 100 | 155 100 | 2500 1600 | 2500 1600 | |
| Whale-liver concentrate | | | | 60 | 130-180 | 180 | 2800-3000 | 3000 | |
| Whale and fish-liver concentrates in vegetable oil: | | | | | | | | | |
| (i) direct | | | | 3.6 | 9.4 | 10.9 | 165 | 175 | |
| (ii) <i>via</i> unsap. | | | | 3.2-3.5 | 9.5-10 | 10-10.9 | — | — | |
| Cod-liver oils | | | | { 1.1 0.5 | 1.5* 0.6* | (3.4) (1.5) | 20 8 | (55) (25) | |
| Same cod-liver oils <i>via</i> unsap. .. | | | | { 0.80 0.32 | 2.4 0.95 | 2.48 1.0 | 40 15 | 40 16 | |
| Two abnormal liver oils | | | | { 14 56 | 28 107 | 43.5 174 | 650 1700 | 700 2800 | |
| Margarine: | | | | | | | | | |
| (i) on fat itself | | | | >0.1 | <0.01 | >0.3 | — | >5 | |
| (ii) <i>via</i> unsap. | | | | >0.03 | 0.03 | >0.1 | ca. 0.5 | >1.5 | |

"Pure" vitamin A ($\text{C}_{30}\text{H}_{48}\text{O}$)—approximate data from which vitamin A content can be calculated:

$E_{1\text{cm}}^{1\%}$ 325m μ , 2000; $E_{1\text{cm}}^{1\%}$ 620m μ , 6200; $E_{1\text{cm}}^{1\%}$ 580m μ , 3100; L-620m μ Value,⁴ 4800; Blue Value, 100,000; Moore Blue Units per g., $5\frac{1}{2}$ million; Dilution Strength⁵, 7-10 million.

* Maximum 603-606m μ .

To "unmask" the vitamin A in the original mixture, saponification is the best method. The following technique, based on S.P.A. procedure,¹² is rapid and reliable, and equally applicable to margarine concentrates and to fish oils of the cod-liver oil type, with the reservation that unaccountable losses of 10-15 per cent. may occasionally occur, despite all precautions:

Dissolve 0.5 to 2.0 g., according to expected potency, in 5 to 10 ml. of cyclohexane, and test as above, thus obtaining useful preliminary and confirmatory information. Boil duplicate 1-ml. portions with 0.2 to 0.4 ml. of 60 per cent. potassium hydroxide solution and 5 ml. of pure alcohol for 5 to 10 minutes (without porous pot, etc.), dilute with 10 ml. of water, extract twice with 25 to 30 ml. of *freshly distilled*⁸ ether, wash combined extracts with 10 ml. of water, 10 ml. of dilute potassium hydroxide solution,† 3 × 10 ml. of water, all at 30° C. Remove ether. Add a few drops of pure alcohol to the wet residue, and blow until dry two or three times with nitrogen or carbon dioxide at 100° C. Avoid strong sunlight and maintain anaerobic conditions at all stages; the saponification mixture must not boil dry, for example, but there is no need for a reflux condenser. Dissolve the dry unsaponifiable matter at once in 5 ml. of cyclohexane and proceed as before with colour and ultra-violet tests, calculating the results as if the original oil were in the solution.

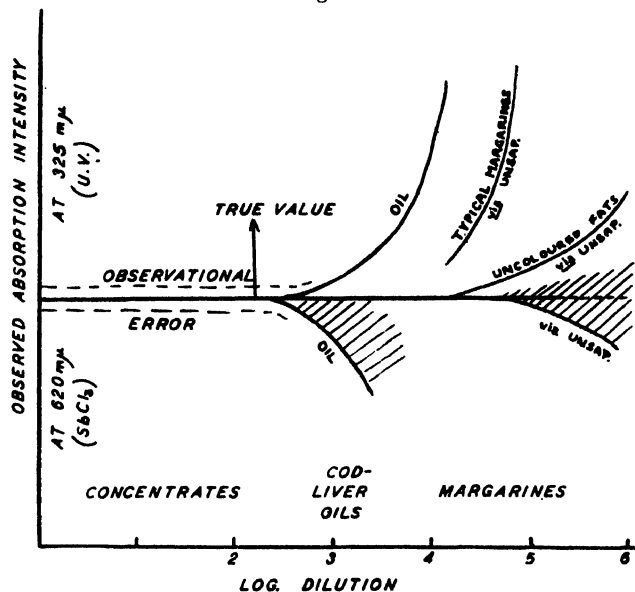
EXAMINATION OF MARGARINES.—While for concentrates the ultra-violet absorption provides the most reliable single criterion of potency, for margarines this method of testing is, in the present state of knowledge, useless owing to the preponderant effect of irrelevant absorption (Table II, Fig. 3). Results may be anything up to several hundred per cent. too high.

† Optional with less than 0.2 g. of oil.

For margarines, the most trustworthy test is the measurement of the absorption at $620m\mu$ on adding antimony trichloride reagent to a solution of the unsaponifiable matter in cyclohexane or chloroform. A convenient procedure is as follows:

From 8 to 10 g. of margarine are boiled for 10 to 15 minutes with 30 ml. of alcohol containing 4 ml. of 60 per cent. potassium hydroxide solution. The mixture is diluted with 60 to 80 ml. of water and extracted three or four times with 50 ml. of freshly distilled ether. The combined extracts are washed with 50 ml. of water, 50 ml. of dilute potassium hydroxide or carbonate solution, and three times with 50 ml. of water, all at 30°C . As a precaution it may sometimes be advisable to re-extract the washings, in view of the low initial potency, which leaves no margin for possible loss. After removal of the ether (without porous pot, etc.), a few drops of alcohol are added and

Fig. 3



Diagrammatic representation of the effect of the diluent oil or fat on $E_{1\text{ cm}}^{1\%}$ $325m\mu$ and $E_{1\text{ cm}}^{1\%}$ $620m\mu$ values, showing how saponification postpones deviation from the truth, and how at high dilutions the antimony trichloride reaction becomes more reliable than the ultra-violet absorption.

the wet residue is blown until dry two or three times at 100°C . under nitrogen or carbon dioxide. The dried unsaponifiable matter is immediately made up to 2 ml. with alcohol-free dry chloroform and the $E_{1\text{ cm}}^{1\%}$ $616-620m\mu$ value is determined as if the solution contained 400-500 per cent. (w/v) of margarine, the intensities being matched as before at as near an optical density ($\log I_0/I$) of 2.0 as possible.

For a quick, though less accurate, test, the ethereal extractions may be reduced to two, washing confined to one portion of 50 ml. of water, and the ether dried with sodium sulphate. Such a test will detect gross irregularities and can be carried through in half an hour.

At its best, the test amounts in practice to a determination of the final dilution required to produce a standard effect, and proportionality is not stretched too far. In this respect the method resembles the "dilution test," but has the advantage that bleaching with charcoal can be avoided. We have found it exceedingly difficult to prevent a loss of 10 to 15 per cent. of the total vitamin A as a result of even the most cautious bleaching. The reaction colour of the unbleached material will probably not be blue, owing to the antimony trichloride reacting with the dye to give a relatively stable red complex; but as a rule the normal $620m\mu$ band is sufficiently separated from other absorption to make estimation to about ± 15 per cent. possible; carotenoids (up to more than the vitamin content) can be ignored.

As a refinement, a second reading may be made at $620m\mu$ after the vitamin A band has faded, and the residual stable absorption, due to the dye and antimony complex, deducted from the gross absorption, due to dye plus vitamin. This correction, actually a slight over-correction, rarely exceeds 10 to 20 per cent. and provides a lower limit to the true value.

LIMITATIONS TO ESTIMATION OF POTENCY.—To these general indications of the technique recommended in the particular cases cited, it will be well to add some brief notes on the interpretation of the results. Limitations to accurate prediction of potency are imposed from three different directions:—(1) The ordinary errors associated with sampling, manipulation, and measurement. (2) Restrictions inherent in the character of the materials under examination. (3) Difficulties arising from the interpretation of the data, especially in relating data to biological activity.

It is not necessary here to insist on the importance of the first of these limitations, but it seems desirable to deal more fully with the other two. Reproducible results are not necessarily correct results. The most important of the limitations inherent in the materials themselves are:—(a) inhibition of colour reaction; (b) enhancement of colour reaction; (c) irrelevant ultra-violet absorption.

(a) The inhibition effect¹¹ is probably best seen in the ordinary blue-value determination on cod-liver oil, and is best overcome by saponifying and making the test on the unsaponifiable matter. Destruction of inhibitors by, for example, bromine, is not always satisfactory. Some inhibition must be expected with many of the margarine concentrates available, but its percentage effect becomes very much less marked with more highly potent preparations and undetectable in the average unsaponifiable fraction. The possibility of occasional unexplained losses on saponification, however, makes it advisable to test all materials directly whether they are also studied *via* the unsaponifiable matter or not.

Partial oxidation of an oil will lead at times to a modification of the blue colour by the introduction of a red constituent, and a quantitative visual comparison is then almost impossible; here again saponification usually overcomes the difficulty.

(b) Enhancement of blue colour may arise from the presence of cyclised or "spurious" vitamin A,¹³ of vitamin A₂,¹⁴ or of a constituent of whale oil, to which attention has been called.^{15,16} With the exception of the last, these may safely be ignored in margarines and margarine concentrates, while the whale constituent fortunately does not sensibly affect the BV/UV ratio if the reagent is iron-free, and so is automatically allowed for by a lower conversion factor (below).

(c) Irrelevant absorption is of special importance in ultra-violet measurements, particularly when the Hilger vitameter or similar instruments are used, since obviously no distinction can be made between absorption at $325m\mu$ due, respectively, to vitamin A and to other ingredients of the preparations. It is clearly important to know how much of the observed absorption is not due to vitamin A. The persistence or crest-to-trough depth of the absorption band provides a useful indication at a glance. Except with whale products, the irrelevant absorption likely to be encountered in most oils, and in margarine concentrates in particular, can be ascribed mainly to glycerides.* The whale-liver oil ingredient—which has also been seen in minor quantities in fish oils—cannot conveniently be removed. Its effect on the absorption spectrum is to displace the maximum from $325m\mu$ towards $290m\mu$ ($315m\mu$ is a common figure), but intensity readings are still made at $325m\mu$ for calculation of potency. Although spectroscopically repugnant, this serves remarkably well in practice. Vitamin A₂ and "spurious" vitamin A together seldom contribute more than 5 per cent. to the total $E_{1\%}^{1cm}$ $325m\mu$ values. When they do, they are fortunately readily detected, and the material must be classified

* This amounts in fish oils to a fairly constant 0.1 to 0.3, a significant proportion with cod-liver oil (total absorption 0.5 to 1.5), but negligible in relation to, say, halibut oil.

as unsuitable for simple physical assay. Glycerides can be either allowed for as already described or, more generally, removed by saponification.

At the potency level of butter and margarines, even the residual absorption by other constituents of the unsaponifiable matter forms a serious proportion of the total absorption; the absorption not ascribable to vitamin A may in fact greatly exceed that due to the vitamin itself (Table II).

TABLE II
PERCENTAGE EFFECT OF IRRELEVANT ULTRA-VIOLET ABSORPTION
(Excluding A₂, spurious A, etc.)

| | E _{1cm.} ^{1%} 325mμ | Per cent. irrelevant absorption | |
|-------------------------------------|---------------------------------------|------------------------------------|------------|
| | | Direct | Via unsp. |
| Halibut, shark, etc., oils | 50 | 0.2-0.6 | negligible |
| 5000 I.U. per g. concentrates | 3.5 | 1-4 | „ |
| Cod-liver oils | 0.5-1.5 | 6-50 | „ |
| Margarine | about 0.1-0.2 | >90 | up to 90 |

It is true that good results have been obtained with some natural rich summer butters by an ingenious modified technique,¹⁷ but this cannot be applied to margarines (or many butters) in which a variable proportion of the non-carotenoid colouring matter remains after saponification and cannot by any means at present known be accurately allowed for, nor completely removed without loss of undefined quantities of vitamin A. Measurement of intensity before and after destruction of the vitamin by irradiation, etc., has been suggested; the effect on other constituents, particularly carotenoids, is, however, unpredictable. Possibly quantitative chromatography will provide an ultimate solution. Meanwhile, only the colour test remains as a practical alternative to the biological assay of margarine.

INTERPRETATION OF DATA.—Conversion of physical data to biological potency, expressed in terms of International Units, is accomplished by the use of suitable conversion factors. Those now used in this laboratory for converting E_{1cm.}^{1%} 325mμ values into I.U. per gram are:—(a) for fish-liver and visceral products: 1600; (b) for refined whale-liver products: 1200.

The first is the figure recommended by the International Conference on Vitamin Standardisation, 1934. The second is based on a large number of assays carried out in this department.

Concurrently with the ultra-violet factors, corresponding factors for the colour reaction receive automatic, if tacit, sanction (Table III). Whether the application

TABLE III
CONVERSION FACTORS—PHYSICAL DATA TO INTERNATIONAL UNITS

| | Fish products I.U. per g. | Refined whale products I.U. per g. |
|---|------------------------------|--|
| E _{1cm.} ^{1%} 325mμ | 1600 | 1200 |
| E _{1cm.} ^{1%} 620mμ | 510 | 380 |
| L-620mμ value ⁴ | 660 | 495 |
| Carr-Price Blue Value | 32 | 24 |
| Moore Blue Units per g... .. | 0.6 | 0.45 |
| Dilution Strength | 0.3-0.5 | 0.24-0.36 |

of any factor is justified depends on whether the material under test can be regarded as spectroscopically normal. Unless the various data conform reasonably (± 10 per cent.) with the ratios for "pure" vitamin A (Table I), the material is abnormal and interpretation of results becomes open to question. Abnormal

materials, if spectroscopically similar, may, of course, legitimately be compared with each other, but not directly with vitamin A.

For all current margarines the appropriate factor for the $E_{1\%}^{1\text{cm}}$ 620m μ value may be taken as 500 without exceeding the limits of error. It would be well, however, to avoid possible confusion with directly obtained biological data by referring to all potencies derived through conversion factors specifically as *Spectroscopic International Units*.

I wish to express my thanks to the Directors of Lever Brothers & Unilever, Ltd., for permission to publish this paper.

REFERENCES

1. F. H. Carr and E. A. Price, *Biochem. J.*, 1926, **20**, 497.
2. T. Moore, *id.*, 1929, **23**, 1267; 1937, **31**, 155.
3. A. Andersen and E. Nightingale, *J. Soc. Chem. Ind.*, 1929, 139r.
4. W. J. Dann and K. A. Evelyn, *Biochem. J.*, 1938, **32**, 1008.
5. Adam Hilger, Ltd., London.
6. R. L. McFarlan, J. W. Reddie and E. C. Merrill, *Ind. Eng. Chem., Anal. Ed.*, 1937, **9**, 324.
7. O. Notevarp, *Biochem. J.*, 1935, **29**, 1227.
8. E. M. Hume and H. Chick, M.R.C. Spec. Rep. No. 202 (1935).
9. E. L. Smith, B. Stern and F. E. Young, *Nature*, 1938, **141**, 551.
10. I. M. Heilbron, A. E. Gillam and R. A. Morton, *Biochem. J.*, 1931, **25**, 1352.
11. R. A. Morton, A. E. Gillam, I. M. Heilbron, J. A. Lovern, M. van Eekelen, A. Emmerie, H. W. Julius, L. K. Wolff; several papers, 1931-33.
12. "S.P.A. Method for the Determination of Unsaponifiable Matter in Oils and Fats," *ANALYST*, 1933, **53**, 203.
13. K. C. D. Hickman, N. D. Embree, etc., several papers, 1935 onwards.
14. J. R. Edisbury, R. A. Morton, G. W. Simpkins and J. A. Lovern, *Biochem. J.*, 1938, **32**, 118; *Nature*, 1938, **140**, 234.
15. H. Pritchard, H. Wilkinson, J. R. Edisbury and R. A. Morton, *Biochem. J.*, 1937, **31**, 258.
16. R. T. M. Haines and J. C. Drummond, *ANALYST*, 1938, **63**, 335.
17. A. E. Gillam, I. M. Heilbron and R. A. Morton; several papers from 1933, *cf.* particularly *Biochem. J.*, **28**, 79.

CENTRAL TECHNICAL DEPARTMENT/FOOD
LEVER BROTHERS & UNILEVER, LTD.
PORT SUNLIGHT, CHESHIRE

May 17th. 1940

DISCUSSION

Dr. F. H. CARR asked if Dr. Edisbury could give any figures comparing the dilution test with what he himself must have known to be the truth from biological tests. (Now published in Table III, p. 491.) If it would stand the test of comparison the dilution test would surely be of very great use to analysts. In Dr. Carr's view, the u.v. conversion factor of 1200 for whale oil, and even that of 1600 for vitamin A, must be the subject of controversy. While 1600 was the figure adopted by the International Committee on Vitamin Standards, in U.S.A. the factor for pure vitamin A was thought to be over 2000.

Mr. A. L. BACHARACH said that Dr. Edisbury had several times referred to whale oil concentrates. He took it that Dr. Edisbury meant products made from whale liver oil and not whale body oil; it seemed to him particularly desirable at the present time to make the distinction clear.

Mr. J. R. NICHOLS asked if Dr. Edisbury had ever been unable to get a dilution value on margarines? With certain margarines he himself had found a fading of colour, leaving finally nothing but a pinkish shade. The dilution might be 1 in 5 or 1 in 50, and one could not tell the difference. Could the author say where he had gone wrong in getting such a result with margarine?

Dr. EDISBURY, replying, said that, in his opinion, justification for the use of conversion factors turned on whether vitamin A was a single substance or a mixture of isomers, etc., spectroscopically similar, but biologically different. If heterogeneity was involved, one certainly got remarkably constant mixtures from a variety of sources, and the ultra-violet conversion factors recommended for fish and whale products were, with a few exceptions, regularly obtained experimentally. Assuming a unique vitamin A with $E_{1\%}^{1\text{cm}}$ 325m μ 2000, and defining the International Unit as the activity of 0.6 γ of β -carotene, any factor over 800 implied inefficient utilisation of the carotene standard by the test animals, while 1600 indicated the (fortunately consistent) effective use of one molecule of vitamin A out of the theoretically possible two from each molecule of β -carotene. He wondered whether the higher factors used in America originated through hyper-activity of the U.S.P. Reference Oil, or less than 50 per cent. efficient utilisation of carotene, when this subsidiary standard was initially "calibrated." American

factors were further exaggerated by ignoring the demonstrable deterioration of the U.S.P. oil. High factors were, of course, immensely popular with vendors. As regards the other questions, all whale products referred to were liver concentrates. Finally, the unsatisfactory colour obtained in margarine-testing was the chief reason for recommending the spectroscopic test, which, by measuring absorption intensity at one wavelength, avoided any question of matching dissimilar colours. In the simpler dilution test success depended on first bleaching out the dye. Fading was inevitable in either test; hence the need for rapidity.

Estimation of Vitamin D in Margarine

BY N. T. GRIDGEMAN, B.Sc., H. LEES, B.Sc., AND H. WILKINSON, Ph.D.

(Read at the Meeting, May 1, 1940)

IT is now compulsory that all margarines sold in this country be vitaminised with vitamins A and D. The estimation of vitamin A by spectrophotometric methods has been discussed by Edisbury¹ (p. 484); in this paper we deal with the estimation of vitamin D.

There have been many chemical tests proposed for the estimation of vitamin D, but interference by other substances such as sterols, vitamin A and carotenoids, has so far made it impossible to apply them to margarine. Moreover, the potency of margarine (about 1 I.U. of vitamin D per gram—equivalent approximately to 0.025 p.p.m.) is very much lower than any which other investigators have estimated colorimetrically.

It is necessary therefore to use a biological method. There are three in general use, and all depend upon assessment of the antirachitic activity of vitamin D. They are the bone-ash method, the X-ray method and the line-test method. The first is prophylactic and depends upon assessing the degree of prevention, by vitamin-D administration, of the normal fall in the bone ash-content in rats weaned on to, and maintained on, a rachitogenic diet for a test period of 3 to 5 weeks. The other two methods are curative tests carried out on animals in which rickets has first been produced by a rachitogenic diet. In the X-ray method, the healing due to administration of vitamin D is assessed radiographically. In the last method, the line test, the healing that takes place between the cap and the shaft of the radius and ulna is also evaluated, but the assessment is dependent upon visual or photographic examination of stained sections of the bones.

In the assay of margarine a modification of the line-test method has been used in this laboratory for the last 7 years. The statistical error of this test has been calculated to be 76 per cent. to 132 per cent. ($P = 0.99$) for ten pairs of rats. The technique, in which camera-lucida drawings of the stained distal ends of sagittal sections of the radii and ulnae are measured by a planimeter, was described by Morgan.⁴ The diet given by him has been modified (Wilkinson⁵) to maize (flaked and ground), 84; meat meal, 12; salts, 4 per cent. The essence of the test consists in comparing the extent of healing of rickets effected over a ten-day period by daily doses of 0.25 or 0.5 I.U. of vitamin D Standard per rat, with that effected in a second group of rats receiving equivalent doses of the vitamin-containing material.

It is necessary that this material should be dosed to the rats in quantities or dilutions containing as nearly as possible either 0.25 or 0.5 I.U. even if, with a material of unexpectedly high or low potency, a re-assay has to be carried out. Appropriate doses of a margarine of unusually low potency, say of the order of 0.1 I.U. per gram, will constitute a significant fraction of the daily food-intake, and in consequence the consumption of the diet itself will be reduced to an extent detrimental to the healing mechanism. Lines on which a modified technique—involving a compensatory increase of the rachitogenic salt-mixture in the diet of the "margarine" rats—could be developed to meet such a contingency, have been

suggested elsewhere.² Extraction and assaying of the unsaponifiable fraction of the margarine fat should theoretically provide another solution, but in practice it has been found difficult to avoid vitamin-D losses during the chemical manipulation. Presumably margarine vitaminised much below normal will be encountered only infrequently, and therefore this necessarily tedious—and expensive—method of arriving at a reasonably accurate assay, will not often be called for. Nevertheless it would obviously be advantageous to have a vitamin-D test which could more easily be applied to margarines of all likely potencies.

This consideration, among others, led us to examine the merits of the bone-ash technique. This was before Henry and Kon³ reported results, obtained by this method, on the antirachitic action of fat. While this may introduce a difficulty

TABLE I

| Substance | Line-test ratio | Bone-ash test ratio |
|-----------|-----------------|---------------------|
| A | 108 | 96 |
| | | 96 |
| B | 110 | 98 |
| | | 97 |
| C | 76 | 71 |
| D | 103 | 93 |
| E | 87 | 78 |
| F | 108 | 97 |
| G | 97 | 85 |
| | | 88 |
| H | 69 | 50 |
| | | 49 |
| J | 83 | 68 |
| | | 68 |

into assaying material of low potency, we ourselves have encountered a further difficulty, which applies generally under the conditions of our experiments.

The bone-ash technique in use in this laboratory is based on the normal prophylactic technique described in the literature. The rachitogenic diet given above is employed during a test period of 3 weeks from the day of weaning. Dosage levels of 0.1 and 0.2 I.U. per day are generally used, although even lower levels can be used if a 5-week period is adopted. The ash content of the fat-free moisture-free tibia and femur is determined and used as an inverse measure of the rickets. A number of antirachitic substances have been assayed by both methods (line test and bone-ash) and five repeat assays have been performed. The results are expressed as the ratio:

$$\frac{\text{Potency of Test Substance Dose}}{\text{Potency of International Standard Dose}} \times 100,$$

and are given in Table I. The statistical errors of the ratios are not given.

The errors of both sets of tests were about the same. The results show that the bone-ash method allows very good repetition, possibly better, in fact, than the line-test method. However, the most notable feature is that in all the bone-ash assays the potency, expressed in the Table as a ratio, is lower by about 10 per cent. The materials which form the subject of the above Table cover a large range of potencies from 0.5 I.U. to 40×10^6 I.U. per gram.

In seeking to explain these differences we must bear in mind that the line-test measures a purely localised reaction, while the bone-ash test measures the results of a more generalised one. Throughout all this work the International Standard has been used. If the International Standard had been calciferol, pure vitamin D₂, which is one of the test substances included in Table I, then the assays obtained

by line-test and bone-ash methods would have been identical. It is possible that there is, in the International Standard, which is the only crude irradiated material used in this work, some substance that produces a greater response in the bone ash than in the line test. Two experiments have been carried out to examine this possibility.

The line test is necessarily curative and the bone-ash method is usually prophylactic, but by means of a prolonged line test it has been possible to carry out on the same animals both line-test and curative bone-ash assays. The results are given in Table II.

TABLE II
CURATIVE LINE-TEST AND BONE-ASH ASSAYS ON THE SAME ANIMALS

| Expt. | Ratio: $\frac{\text{Potency of test substance dose}}{\text{Potency of International Standard dose}} \times 100$ | | $\frac{\text{Line test ratio}}{\text{Bone-ash ratio}}$ |
|-------|--|---------------|--|
| | Line test | Bone-ash test | |
| 1 | 122 | 102 | 1.2 |
| 2 | 165 | 125 | 1.3 |

The results show that the ratio between the effects of two substances, one of which is the International Standard, is different for the line-test and for the bone-ash test. This indicates that the crude irradiated material constituting the International Standard has two different effects, according as it is used in the line-test or the bone-ash method.

SUMMARY.—Margarines of potency 1.0 International Units of vitamin D per gram can be assayed satisfactorily by the normal line-test method. Assays of the vitamin concentrates and the margarine made with them show that the margarine assay is not falsified by the obligatory feeding of fat to the test-animals. The assay of very low-potency margarines, however, entails the feeding of larger quantities of fat, which have been found to affect the responses of the test-animals. The more sensitive bone-ash assay has been suggested as a preferable method. This has been tested, but the two methods give somewhat different results. It is possible that the International Standard is not so effective in the line-test as it is in the bone-ash assay. Results indicating such a difference in activity have been obtained.

We wish to thank the Directors of Lever Brothers & Unilever, Ltd., for permission to publish these results.

REFERENCES

1. J. R. Edisbury, *ANALYST*, 1940, **65**, 484.
2. N. T. Gridgeman, H. Lees and H. Wilkinson, *Biochem. J.*, 1939, **33**, 641.
3. K. M. Henry and S. K. Kon, *Chem. and Ind.*, 1940, **59**, 184.
4. R. S. Morgan, *Biochem. J.*, 1932, **26**, 1144.
5. H. Wilkinson, *ANALYST*, 1939, **64**, 17.

CENTRAL TECHNICAL DEPARTMENT/FOOD
LEVER BROTHERS & UNILEVER, LTD.
PORT SUNLIGHT, CHESHIRE

DISCUSSION

MR. A. L. BACHARACH said that the paper threw a good deal of light on a very tricky problem. He thought that the suggested explanation of the fact that two different methods of assay gave significantly different results for the amount of vitamin D, in international units, present in oils, concentrates and calciferol, was probably the correct one. Nobody was really to blame for the fact that the International Standard Preparation behaved somewhat differently from pure calciferol; at the time the Preparation was made available, pure calciferol was not known. The authors' results differed from those of Dr. S. K. Kon in that they were unable to detect any antirachitic action of fat as such. This was fortunate, because it meant that they could feed their test sample direct to the animals without previous saponification to remove glycerides, and

this would obviously involve a considerable saving of time and would also avoid the risk of losing vitamin D.

Dr. H. E. Cox remarked that a few years ago Brockmann and Chen published a modification of the antimony chloride test which was supposed to give a quantitative determination of vitamin D. It was now known that as a quantitative test the method failed, but he would like to know whether the authors considered it of any qualitative value; if it could be used, it would be so much simpler than bone tests.

Dr. WILKINSON, replying, said that all he had wanted was a reliable method for the assay of margarine. Dr. Kon had used starch in his diet and had not added vitamin D, and therefore his experiments were not really comparable with those previously reported by Lees, Gridgeman and Wilkinson. Replying to Dr. Cox, he said that the spectrophotometric absorption due to one unit per gram would be so small that it would not be detectable in the test. In his laboratory they had not been able to apply the Brockmann-Chen test to margarine or margarine concentrates.

The Determination of Phosphorus in Titanium Steels*

By A. T. ETHERIDGE, M.B.E., B.Sc., F.I.C., AND D. G. HIGGS

IN a paper read before the Cleveland Institution of Engineers in 1920, Ridsdale¹ drew attention to the inaccurate results for phosphorus in cast iron caused by the presence of 0.1 to 0.4 per cent. of titanium. Apparently the low results obtained are due to the phosphorus being incompletely precipitated as phosphomolybdate. No explanation of the cause was suggested, but it seems probable that titanium may form a complex compound with the PO_4''' anion, thus competing with the formation of the molybdate complex. To prevent this interference Ridsdale put forward two remedies: (1) removal of titanium by cupferron precipitation; (2) modification of reagents, involving a double precipitation of the phosphomolybdate. Both methods appear to be accurate but are, of course, more cumbersome than the usual procedure for determining phosphorus in steel.

The matter has not received much attention in text-books apart from the statement that titanium, if present in quantity, must be removed before phosphorus is determined, but no attempt has been made to formulate the maximum percentage of titanium permissible for the accurate determination of phosphorus. This is probably due to the fact that titanium is rarely found in steel except in minute traces, which admittedly have no effect. Modern steel research, however, is gradually including the testing of alloys formed with elements not previously tried, and titanium is one of these. It has therefore become necessary to find a reliable method of determining phosphorus by a single precipitation without removing titanium. Preliminary work established the fact that the maximum amount of titanium consistent with this requirement is approximately 1 per cent. When the titanium-content exceeds that amount it cannot be held in solution in the usual nitric acid solvent (sp.gr. 1.2), so that under such conditions separation at the outset by a method on the lines described by Ridsdale (*loc. cit.*) appears to be the only practicable course. Considerable interest attaches, however, to the effect on steel of titanium of the order of 1 per cent. or less.

A series of tests was carried out on electrolytic iron (containing a mere trace of phosphorus, approximately 0.003 per cent.) to which were added known amounts of titanium and phosphorus. The standard titanium solution was prepared from potassium titanium oxalate, the oxalate radical being decomposed by boiling with conc. nitric acid, evaporating the solution to a small bulk and diluting with nitric acid (sp.gr. 1.2) to a known volume. The titanium-content of this solution was checked gravimetrically in the usual way. The standard phosphate solution was prepared from AnalaR sodium phosphate, a calculated amount being weighed

* A communication from the Research Department, Royal Arsenal, Woolwich.

and dissolved in nitric acid (sp.gr. 1.2) and diluted to a known volume with the same acid. The use of nitric acid of that strength as a solvent and diluent facilitates the preparation of the tests which are described below. For each test the usual two-gram weight of electrolytic iron was used.

In this first series the method described by Etheridge² was followed and the results are shown in Table I, Series I.

TABLE I

| Titanium added Per Cent. | Phosphorus added Per Cent. | Phosphorus found* | | |
|--------------------------------|----------------------------------|-----------------------|------------------------|-------------------------|
| | | Series I Per Cent. | Series II Per Cent. | Series III Per Cent. |
| 1.00 | 0.020 | 0.011 | 0.018 | 0.021 |
| 1.00 | 0.030 | 0.025 | 0.029 | 0.030 |
| 1.00 | 0.040 | 0.032 | 0.035 | 0.041 |
| 1.00 | 0.050 | 0.042 | 0.045 | 0.051 |
| 1.00 | 0.060 | 0.048 | 0.055 | 0.061 |
| 1.00 | 0.070 | 0.059 | 0.065 | 0.070 |
| 1.00 | 0.080 | 0.068 | 0.073 | 0.079 |
| 1.00 | 0.090 | 0.074 | 0.084 | 0.091 |
| 1.00 | 0.100 | 0.083 | 0.096 | 0.100 |
| 0.50 | 0.040 | — | 0.039 | — |
| 0.50 | 0.060 | — | 0.057 | — |
| 0.50 | 0.080 | — | 0.077 | — |
| 0.25 | 0.040 | — | 0.039 | — |
| 0.25 | 0.060 | — | 0.058 | — |
| 0.25 | 0.080 | — | 0.078 | — |
| 0.10 | 0.040 | — | 0.039 | — |
| 0.10 | 0.060 | — | 0.059 | — |
| 0.10 | 0.080 | — | 0.080 | — |

* Corrected for phosphorus in the electrolytic iron used (0.003 per cent.).

It is obvious that the results thus obtained are low. It was suggested (by W. J. Agnew, of this Department) that vigorous stirring with a "policeman" rod immediately after the addition of the molybdate reagent, should be tried, this having been found beneficial in the somewhat similar interference of vanadium (*vide infra*). For this purpose tests were carried out in tall beakers to make possible the vigorous stirring referred to above. The results obtained are shown in Table I, Series II.

These results show that (1) the modification of vigorous stirring gives better results; (2) as expected, the results improve with diminishing titanium; (3) correct results cannot be obtained if the titanium exceeds 0.1 per cent.

At this point it occurred to us that Johnson's process³ for vanadium steels, as modified in this Department, might also be effective for titanium steels. This process was originally put forward as a remedy for the well-known interference of vanadium. It requires the presence of nitric acid in much larger excess and of greater concentration than in the customary methods. Johnson operates on 1.63 g. of steel and requires the solution to stand overnight. Agnew has modified this process by using the usual two-gram. weight of steel and stirring vigorously after adding molybdate as previously described. Under these conditions it is unnecessary for the solution to stand overnight; on the contrary, the determination can proceed after the usual period of standing. Correct results have been obtained with vanadium steels by this method.

This modified process was used in a third series of tests and, as is shown in the last column of Table I, it gives correct results.

It is carried out as follows:—Two g. of steel are dissolved in 45 ml. of nitric acid (sp.gr. 1.2) in a tall beaker, digested if necessary (to decompose carbides) and oxidized with permanganate solution, the excess of which is destroyed with sodium or potassium nitrite as described by Etheridge.⁴ Fifty ml. of conc. nitric acid

(sp.gr. 1.42) are added and the solution is boiled for one minute with a glass cover on the beaker. The beaker is removed from the hot plate, the cover and sides are rinsed lightly with hot water, 50 ml. of cold molybdate reagent* are added, and the mixture is vigorously stirred for some minutes with a "policeman" rod. After standing for approximately 30 minutes the precipitate is filtered off and dealt with as usual.

REFERENCES

1. N. D. Ridsdale, *Proc. Cleveland Inst. Engineers*, Feb. 9th, 1920.
2. A. T. Etheridge, *ANALYST*, 1931, **56**, 14.
3. C. M. Johnson, "*Rapid Methods for the Chemical Analysis of Special Steels*," 4th Ed., 1930. March 18th, 1940

The Quantitative Electro-deposition of Tin from Chloride Solutions

Part I. Stannic Tin Solutions

By F. G. KNY-JONES, M.Sc., A. J. LINDSEY, M.Sc., Ph.D., A.I.C., AND
A. C. PENNEY, M.Sc., A.I.C.

ADVANTAGES of using hydrochloric acid solutions for rapid electrolytic separation of metals have been set forth in previous communications from this laboratory.^{1,2,3,4} The present paper describes a study of the conditions necessary for the quantitative deposition of tin in this way. Several analysts^{5,6,7} have been unable to remove residual quantities (up to several mg.) of tin during electro-deposition from chloride solution, but no adequate explanation has been given; errors of similar magnitude can be found in the work of others. It has been suggested that small quantities of tin may be lost: (1) by formation of gaseous hydride at the cathode, (2) by volatilisation of stannic chloride during the preparation of the solution, (3) by re-solution of the deposit during washing, and (4) by mechanical loss of tin crystals from deposits of poor quality. We deal with these suggestions in turn.

Formation of Stannane.—This may occur to the extent of 0.01 per cent. of the evolved gases obtained by electrolysing a solution of tin sulphate between lead electrodes.⁸ Schleicher and Toussaint⁹ could not detect tin hydride under the conditions of electro-analysis, and this is in accord with the observation of Paneth and Rabinowitsch⁸ that stannane is decomposed in presence of either tin or strong mineral acids.

Volatilisation of Stannic Chloride.—In our experience this does not occur in the preparation of solutions for analysis provided prolonged boiling with acid is avoided. For example, when a mixture of 2 g. of stannous chloride, 0.5 g. of potassium chlorate and 15 ml. of conc. hydrochloric acid was distilled in an all-glass apparatus the first 1 ml. of distillate contained no tin detectable with toluene dithiol¹⁰ and the first 5 ml. contained only quantities up to 1 mg. When the same apparatus was used for the preparation of solutions from metallic tin as described below, no tin was volatilised. Addition of ammonium chloride or sodium chloride,¹¹ recommended by some analysts, is therefore unnecessary; moreover, as ammonium chloride promotes re-solution of the deposits (*vide infra*), it is better excluded from the electrolyte.

Solution of Deposit.—There is considerable risk of losing a portion of a tin deposit by re-solution during washing, this loss being promoted by electrolytic action between the tin deposit and the platinum electrode; as was to be expected

* Fifty-five g. of ammonium molybdate and 50 g. of ammonium nitrate, with 40 ml. of ammonia (sp.gr. 0.95) are dissolved in 700 ml. of hot water, and the solution is cooled, made up to one litre, and allowed to stand overnight; it should be filtered before use.

the loss is less when the cathode is coated with a less noble metal such as copper. Although no tin can be detected in the electrolyte after removal of the electrode, considerable amounts may be found in the washings. In our opinion this loss arises in the thin film of electrolyte covering the deposit just before and during washing. Thus in a series of about 25 experiments it was found that when a well-adherent deposit of tin was dipped for ten seconds into the electrolyte from which it had been separated there was a loss of about 1 mg., rising to 1.8 mg. when the temperature was raised from 20° to 40° C. and becoming still greater if the electrolyte made contact with the platinum as well as the deposit. Losses of tin during washing may be largely avoided by the analytical procedure described below.

Mechanical Loss of Deposited Tin.—Much attention has been given to the production of a good film. Large quantities of oxalic acid were recommended by Classen¹² and by Schoch and Brown,⁵ the underlying principle being that with stannic tin the complex oxalate assists the formation of a good deposit by lowering the effective concentration of tin ions. As the same result may be achieved by lowering the cathodic current density, it would appear superfluous to add oxalates unless there is some other reason, such as in the separation of bismuth from tin and lead,⁴ or in the fluoride separation of lead and tin.¹³ It has been noted by Schleicher¹⁴ and Lassieur¹⁵ that the use of oxalate provides an anodic depolariser, but it is recognised by both that further addition of hydroxylamine is desirable to prevent any evolution of chlorine at the anode.

Engelenberg¹⁶ lays stress on the importance of avoiding cathodic evolution of hydrogen, and recommends the use of a persulphate to prevent this. Svěda and Uzel¹⁹ confirm the good effect produced by persulphate in presence of hydroxylamine on the deposit from stannous solutions, but find that larger quantities of hydroxylamine alone have a similar effect. The explanation offered is that hydroxylamine acts as both anodic and cathodic depolariser, being reduced at the cathode to ammonia.

In our investigations we have kept in mind the point that the tin in a solution for analysis may be in either stannous or stannic condition. We have confirmed the reduction of stannic ions during the deposition of antimony or antimony and copper,¹⁸ and find that very little reduction occurs on the platinum electrode when tin alone is being deposited. We consider both hydroxylamine and hydrazine to be suitable anodic depolarisers and that a cathodic depolariser is not necessary provided that cathodic potential control is exercised. We cannot support the statement of Fischer and Schleicher that "hydrazine is unsuitable."¹⁹

PREPARATION OF SOLUTION.—The metal (tin or alloy) is attacked by a mixture of 10 ml. of hydrochloric acid (sp.gr. 1.16) and either 1 ml. of nitric acid (sp.gr. 1.42) or a little solid potassium chlorate, as little heat as possible being applied. When solution is complete a further 5 ml. of hydrochloric acid and the depolariser are added, and the solution is diluted to the required volume for analysis. If metals more noble than tin are first separated the final volume may be as much as 250 ml. We varied the volumes from 100 to 250 ml., the temperature from 20° to 70° C., and the quantity of depolariser from 0.5 to 4.0 g. In all the tests described here hydrazine (as sulphate or hydrochloride) was used as depolariser, but hydroxylamine is equally satisfactory.

In a number of experiments the electrolyte was neutralised just before the end to minimise loss during washing. The electrodes of Sand¹ were used, and all quoted potentials refer to his design of auxiliary saturated calomel electrode. In most of the tests the initial current was between 1.5 and 3 amperes and the electrolysis time for a quantity of 0.3 g. of tin was 30 to 35 minutes. "Chempur" tin was employed for most of the determinations.

Particulars of the determinations made are given in Table I. They show that in absence of ammonium salts the procedure followed gives very accurate results. The presence of ammonium salts gives rise to serious errors, but these can be

avoided by neutralising the solution just before the electrolysis is terminated. Experiments K3 to K6, in which 5 g. of oxalic acid was added to the solution, show that errors caused by this substance can be avoided by keeping the temperature above 40° C. during deposition.

TABLE I

| Expt. | Temp. °C. | Depolar- iser | Volume ml. | Potential volt | Tin taken g. | Tin found g. | Error g. |
|--|--------------|------------------|--|-------------------|-----------------|-----------------|-------------|
| <i>Depositions on platinum electrode.</i> | | | <i>Ammonium salts absent.</i> | | | | |
| K 1 | 30-35 | A 4 g. | 100 | 0.6-0.75 | 0.1505 | 0.1503 | -0.0002 |
| K 2 | 20 | A 4 g. | 250 | 0.6-0.8 | 0.1574 | 0.1576 | +0.0002 |
| K 3 | 20 | A 4 g. | 250 | 0.6-0.8 | 0.1511 | 0.1502 | -0.0009 |
| K 4 | 40-45 | A 4 g. | 250 | 0.6-0.85 | 0.1530 | 0.1531 | +0.0001 |
| K 5 | 40-45 | A 4 g. | 250 | 0.6-0.85 | 0.1535 | 0.1536 | +0.0001 |
| K 6 | 40-45 | A 4 g. | 250 | 0.6-0.85 | 0.1500 | 0.1497 | -0.0003 |
| <i>Depositions on copper-plated electrode.</i> | | | <i>Ammonium chloride (1-2 g.) present.</i> | | | | |
| P 7 | 32 | A 4 g. | 200 | * | 0.2415 | 0.2384 | -0.0031 |
| P 8 | 45 | A 4 g. | 300 | * | 0.2047 | 0.2006 | -0.0041 |
| P 9 | 30-35 | B 4 g. | 200 | * | 0.1342 | 0.1317 | -0.0025 |
| P 10 | 35 | A 4 g. | 200 | * | 0.2788 | 0.2786 | -0.0002 |
| P 11 | 40 | B 4 g. | 200 | 0.7-0.8 | 0.2048 | 0.2051 | +0.0003 N |
| P 12 | 35 | B 4 g. | 200 | * | 0.2055 | 0.2051 | -0.0004 N |
| P 13 | 40 | A 4 g. | 200 | * | 0.3011 | 0.3007 | -0.0004 N |
| P 14 | 40 | A 4 g. | 200 | * | 0.2477 | 0.2472 | -0.0005 N |
| <i>Depositions on copper-plated electrode.</i> | | | <i>Ammonium salts absent.</i> | | | | |
| L 15 | 35-40 | B 4 g. | 100 | 0.6-0.7 | 0.1920 | 0.1919 | -0.0001 |
| P 16 | 35 | B 4 g. | 100 | * | 0.1895 | 0.1888 | -0.0007 |
| P 17 | 30-35 | B 4 g. | 150 | * | 0.1912 | 0.1915 | +0.0003 |
| P 18 | 35 | B 4 g. | 100 | * | 0.1994 | 0.1983 | -0.0011 |
| L 19 | 35-40 | B 4 g. | 100 | 0.65-0.75 | 0.2068 | 0.2069 | +0.0001 |
| P 20 | 35 | B 4 g. | 150 | * | 0.1725 | 0.1727 | +0.0002 N |
| P 21 | 35 | B 4 g. | 150 | * | 0.2442 | 0.2446 | +0.0004 N |
| <i>Depositions on platinum electrode.</i> | | | <i>Ammonium salts present.</i> | | | | |
| K 22 | 35-40 | B 0.5 g. | 100 | 0.6-0.8 | 0.1504 | 0.1486 | -0.0018 |
| K 23 | 35-40 | B 0.5 g. | 100 | 0.6-0.8 | 0.1516 | 0.1502 | -0.0014 |
| K 24 | 70 | B 1 g. | 100 | 0.6-0.8 | 0.2456 | 0.2444 | -0.0012 |
| K 25 | 70 | B 1 g. | 100 | 0.6-0.8 | 0.2503 | 0.2502 | -0.0001 N |
| K 26 | 70 | B 1 g. | 100 | 0.6-0.8 | 0.2427 | 0.2427 | 0.0000 N |
| K 27 | 70 | B 1 g. | 100 | 0.6-0.8 | 0.2514 | 0.2515 | +0.0001 N |

A = Hydrazine sulphate. B = Hydrazine hydrochloride. N denotes that the solution was neutralised at the end of the deposition. * Denotes that the potential was adjusted at the outset and later allowed to rise in order to preserve a constant current.

In Expts. K 3-K 6 inclusive, 5 g. of oxalic acid were added in order to simulate the conditions existing after bismuth has been removed by the method of Kny-Jones.⁴

SUMMARY.—Good results are obtained in the deposition of tin from stannic solutions when potential control is exercised and when ammonium salts are absent, or, if they are present, when the solution is neutralised just before the end of the electrolysis.

We wish to conclude by thanking Dr. Sand for his helpful criticism and advice.

REFERENCES

1. A. J. Lindsey and H. J. S. Sand, *ANALYST*, 1934, **59**, 335.
2. S. Torrance, *id.*, 1937, **62**, 719.
3. A. J. Lindsey, *id.*, 1938, **63**, 159.
4. F. G. Kny-Jones, *id.*, 1939, **64**, 172.
5. E. P. Schoch and D. J. Brown, *J. Amer. Chem. Soc.*, 1916, **38**, 1660.
6. N. H. Furman, *J. Ind. Eng. Chem.*, 1923, **15**, 1071.
7. A. Lassieur, *Compt. rend.*, 1924, **179**, 827.
8. F. Paneth and E. Rabinowitsch, *Ber.*, 1924, **57B**, 1877.

9. A. Schleicher and L. Toussaint, "*Elektroanalytische Schnellmethoden*," Stuttgart, 1926, p. 191.
10. R. E. D. Clark, *ANALYST*, 1936, **61**, 242.
11. T. Grosset, *Bull. Soc. chim. Belg.*, 1933, **42**, 269.
12. A. Classen, "*Quantitativ Analysen durch Elektrolyse*," Berlin, 1920 (6th Edn.), p. 191.
13. A. Lassieur, "*Electroanalyse rapide*," Paris, 1927, p. 116.
14. A. Fischer and A. Schleicher, *id.*, p. 190.
15. A. Lassieur, *id.*, p. 106.
16. A. J. Engelenberg, *Z. anal. Chem.*, 1923, **62**, 257.
17. J. Svěda and R. Uzel, *Coll. Czech. Chem. Comm.*, 1929, **1**, 203.
18. A. Lassieur, *id.*, p. 115.
19. A. Fischer, *id.*, p. 190.

THE SIR JOHN CASS TECHNICAL INSTITUTE
JEWRY STREET
LONDON, E.C.3

April 6th, 1940

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

OBSERVATIONS ON THE USE OF ELECTRODIALYSIS IN THE ANALYSIS OF SOILS

IN testing the applicability of Basu's electrodialysis apparatus¹ to the soils of Palestine, samples were collected (a) from the hilly land at Rehovoth in the south, and (b) from the marshy ground of Hederah in the north. As both soils were homogeneous, the samples were taken at depths of 0 to 120 cm. The first was a red sand soil poor in lime (less than 1.5 per cent.), and the second was a dark heavy soil also poor in lime (less than 0.5 per cent.). Tests for replaceable calcium were made (a) by leaching with *N* ammonium chloride solution (Wright),² and (b) by electrodialysis. Each sample was then mixed with 20 per cent. of calcium carbonate and the replaceable calcium was determined by Hissink's³ method* and by electrodialysis. The results, as mg. equivalents of replaceable calcium per 100 g. of soil, are given in Table I.

TABLE I

| Depth in cm. | Dialysis | Treatment with NH ₄ Cl | Mixed sample containing 20 per cent. of calcium carbonate | |
|-------------------|----------|--------------------------------------|--|---------------------------|
| | | | Hissink's method | Dialysis |
| <i>Light soil</i> | | | | |
| 0-30 | 4.00 | 4.20 | 4.40 | } No end-point reached |
| 30-50 | 4.55 | 4.60 | 4.45 | |
| 50-100 | 4.50 | 4.50 | 4.30 | |
| 100-120 | 4.50 | 4.60 | 4.45 | |
| <i>Heavy soil</i> | | | | |
| 0-30 | 29.80 | 30.00 | 30.20 | } No end-point reached |
| 30-50 | 30.55 | 30.60 | 30.75 | |
| 50-100 | 30.75 | 31.00 | 31.05 | |
| 100-120 | 30.80 | 30.80 | 30.95 | |

These results indicate that whilst Basu's apparatus is of value for determining replaceable bases in Palestinian soils poor in lime, it is inadequate with (artificially prepared) highly calcareous soils.

I wish to thank the Government Analyst, Mr. G. W. Baker, for his advice, and Miss S. Adler for assistance in the analytical work.

REFERENCES

1. J. K. Basu, *J. Agric. Sci.*, 1931, **21**, 484.
2. C. H. Wright, "*A Handbook for Soil Analysis*," London, 1934, pp. 211-214.
3. *Internat. Mitt. Bodenkunde*, 1922, **12**, 81.

GOVERNMENT CENTRAL LABORATORIES
JERUSALEM

M. PUFFELES

February, 1940

* The soil is leached twice with *N* sodium chloride solution and the calcium in each extract is determined; the difference between the results is regarded as exchangeable calcium.

THE DETERMINATION OF BENZOIC ACID

SOME time ago we disagreed with another analyst as to the proportion of benzoic acid in a cordial. Both had followed the method of Monier-Williams (*Reports on Public Health and Medical Subjects*, No. 39, 1927; Abst., ANALYST, 1927, 52, 153, 229), and the only variation was that we had sublimed the benzoic acid for 2 hr. at 180° C., whereas the other analyst had sublimed it at 160° C. for 1 to 1½ hours, following the details given on p. 43 of the Report. Our conditions had been fixed as the result of a few control experiments, and, after further investigation, the other analyst was satisfied that he had failed to sublime the whole of the benzoic acid, and we later reached complete agreement.

Monier-Williams's recoveries of benzoic acid in control experiments were satisfactory, and the different conditions that we use for sublimation are necessitated by small, though apparently important, differences between our hot-air oven and that illustrated in his Report.

In our oven, which was purchased specifically for this work, there is a perforated plate about half way between the top and the bottom, and above this, about 5½ cm. from the top, is a flat solid plate. The thermometer is central and there are four holes for the sublimation tubes. These holes are much too wide for the test-tubes, and are flanged. The tubes are therefore held by well-fitting corks. Each tube is arranged so that the lower 4 cm. is inside the oven, and in this position the bottom of the tube is not in contact with the top plate, but about 1½ cm. above it. The thermometer bulb is placed at the same level and probably records the temperature of sublimation fairly closely. In the illustration of the apparatus used by Monier-Williams it would appear that the tubes are actually resting on a metal plate which, in all probability, is at a somewhat higher temperature than is recorded by the thermometer, and this presumably accounts for our differences.

In a more complete investigation, quantities of about 100 mg. of benzoic acid (A.R. quality) were weighed directly into clean, dry test-tubes, the acid was washed down with a little ether, and the solvent was evaporated with suction as in the Monier-Williams method, to leave residues in the same state as would be obtained by application of the method to a sample containing preservative. Acid-washed and ignited sand was then added, a disc of filter-paper was placed in position, and the sublimations were carried out at different temperatures and for different periods of time. The tubes containing the sublimate were dried and weighed, the benzoic acid was washed off with a little ether, and the tubes were re-weighed. The ethereal solutions were evaporated, the residues were dissolved in neutral 50 per cent. alcohol, and the solutions were titrated with N/10 sodium hydroxide solution which had been standardised against benzoic acid under the same conditions. The results for the gravimetric and volumetric determinations of the sublimed benzoic acid were in close agreement and are not given separately in the table below.

The lower portions of the tubes containing the sand were extracted with ether, and any unsublimed benzoic acid was determined by titration.

TABLE I.

| Temp. of sublimation °C. | Time of sublimation Hours | Benzoic acid taken g. | Sublimate recovery Per Cent. | Recovered from sand Per Cent. |
|--------------------------------|---------------------------------|-----------------------------|------------------------------------|-------------------------------------|
| 160 | 1 | 0.1005 | 44 | 47 |
| | 2 | 0.0945 | 77 | 16 |
| | 3 | 0.0923 | 97.5 | 1.3 |
| 170 | 1 | 0.0954 | 73 | 22 |
| | 2 | 0.0928 | 96 | 0.7 |
| | 3 | 0.1076 | 98 | 0.0 |
| 180 | 1 | 0.0912 | 90 | 5.4 |
| | 2 | 0.0887 | 99 | 0.0 |
| | 3 | 0.0884 | 98 | 0.0 |
| 190 | 1 | 0.0969 | 97 | — |
| | 2 | 0.0935 | 99 | — |
| | 3 | 0.0993 | 98 | — |

The results emphasise the necessity for control experiments in all analytical work. It is quite clear that unless the oven used for sublimation is arranged to agree in all respects with the one described by Monier-Williams, either a higher temperature or a longer time may be required for quantitative recovery of the benzoic acid.

DERBYSHIRE COUNTY COUNCIL LABORATORY
COUNTY OFFICES

ST. MARY'S GATE, DERBY

R. W. SUTTON
O. HITCHEN

June 26th, 1940

Notes from the Reports of Public Analysts

The Editor would be glad to receive the Annual or other Reports of Public Analysts containing matter of special interest to the Society. Notes made from such Reports are submitted to the Publication Committee.

CITY OF SALFORD

ANNUAL REPORT OF THE CITY ANALYST FOR THE YEAR 1939

DOUBLE STRENGTH SELF-RAISING FLOUR.—The bag containing the sample was labelled "double strength." The flour yielded 0.55 per cent. of carbon dioxide, a figure falling within the limits (0.4 to 0.6 per cent.) yielded by ordinary self-raising flours. The packers agreed to omit the words from the label.

OIL OF BITTER ALMONDS.—Eight samples of oil expressed from different types of bitter almonds were examined to ascertain what variations might be expected.

| Type of bitter almond oil | Clouding-point (Evers's modified arachis test) °C. | Bieber (B.P.) test | Iodine value (mean of 2 results) |
|---------------------------|---|-----------------------|--|
| Refined | —1.0 to —1.5 | Very pale yellow | 103.3 |
| Cretan | 0.0 to —0.5 | Very pale buff | 104.4 |
| do. | 0.0 to —0.5 | do. | 104.3 |
| Mogador | 0.0 to —0.5 | Colourless | 100.0 |
| do. | 0.0 to —0.5 | do. | 97.6 |
| Sicilian | —0.5 to —1.0 | do. | 99.1 |
| do. | —0.5 to —1.0 | do. | 99.4 |
| Portuguese .. | 0.0 to —0.5 | Very pale brown | 97.2 |

The yellow to brown colours obtained in the Bieber test were extremely faint and did not in any way resemble the pink colours given by mixtures of apricot kernel and almond oils. The figures for the clouding-point approximated closely to those given by Evers (ANALYST, 1937, 62, 99). It will be observed that three of the samples gave iodine values above the B.P. limits of 95 to 100 for almond oil.

AMMONIA.—Two samples, from different retailers, each contained 15.6 per cent. by weight of NH_3 . Proceedings were instituted and the vendors were fined for selling a poison without being on the List of Sellers under Part II of the Poisons List Confirmation Order, 1935.

ACIDITY OF SOOT DEPOSIT.—The figures for the deposit in the special gauges in various parts of Salford are tabulated. The most noticeable feature is their acid nature, as shown by the pH values of the water collected. The monthly averages were as follows:

| | Salford: Peel Park | Salford: Ladywell Sanatorium | Salford: Drinkwater Park | Marple: Nab Top Sanatorium |
|---------------|-----------------------|------------------------------------|--------------------------------|----------------------------------|
| Acidity | 0.55 | 0.46 | 0.42 | 0.23 |
| pH | 3.9 | 4.1 | 4.1 | 4.4 |

The pH due to carbonic acid in the air would be about 5.5; lower figures indicate an acid, and higher figures an alkaline deposit. Considering that Marple is well in the country and that its general figures are better than those obtained in Salford, its acid rainwater is noteworthy. It shows how widespread may be the drift of acid smoke from the cities. G. H. WALKER

Ministry of Food

STATUTORY RULES AND ORDERS. 1940. No. 11*

Emergency Powers (Defence)

Food

The Feeding Stuffs (Maximum Price Order, 1940). Dated January 6th, 1940

THIS Order comprises 20 Articles with four Schedules fixing the maximum prices for the various feeding stuffs specified.

Article 1 defines the terms used in the Order:

"Compound Cake, Compound Meal or Compound Mixture" means a mixture, etc., containing not less than four of the feeding stuffs mentioned in Part A of the Third Schedule in proportions of not less than 5 per cent. of each, and which contains not less than 2.5 per cent. of oil and 12 per cent. of crude protein; but does not include any concentrate.

"Concentrate" means a feeding stuff which contains not less than four of the feeding stuffs mentioned in Part B of the Third Schedule, and which is manufactured for the purpose of being mixed with other feeding stuffs for the production of a complete ration for feeding to animals and which is properly balanced as to its protein, mineral and vitamin content.

"Flour Mill" means a mill where wheat is milled for human consumption.

"Home produced" in relation to any feeding stuffs means produced in the United Kingdom.

Definitions are also given of "Country flour mill," "distributing dealer," "grower," "importer," "sack," "ton" (= 20 cwt. of 112 imperial lb. each), and "wholesale dealer."

Article 2 provides that no specified feeding stuff shall be sold at a price exceeding the appropriate price (*i.e.* the price set out in the First Schedule, or, for compound cakes, as ascertained in accordance with Article 3). Specified sums may be added for kibbling, grinding, sacks, addition of grains or pulses, or of any meals.

Articles 3 to 11 specify the price per ton in excess of the costs of ingredients and manufacture that may be charged by the manufacturer for compound cake, meal or mixture, and the permissible reductions or increases in the appropriate prices for compound cakes, etc., feeding stuffs specified in the First Schedule, and dried sugar beet pulp.

Article 12 provides for the addition of transport charges to the appropriate price, and Article 13 deals with the apportioning of the cost of sacks. Particulars of all additional charges permitted by the Order must be supplied by the vendor on demand by the buyer in writing (Article 14). A reasonable extra charge may be made for credit (Article 15). Article 16 prohibits fictitious or artificial transactions.

THE FIRST SCHEDULE.—*Part A* tabulates maximum prices for wheat by-products, products from maize, barley, oats, pulses, and other cereals, rice bran, dried grains, etc., and sundry products (fish meal, whale meal, etc.).

Part B includes cotton-seed cake and meal, ground nut cake and meal, linseed cake and other oilseed cakes and meals.

Part C gives maximum prices for dried sugar beet pulp, plain or molassed, ex factories in different parts of England.

Part D gives maximum prices for maize, barley, oats, beans, peas, grain, lentils and locust beans; *Part E* the prices for imported wheat; *Part F* the prices for different kinds of hay, home-produced and imported.

THE SECOND SCHEDULE tabulates the permissible charges per ton additional to the appropriate prices of the specified feeding stuffs, etc., when sold to a person buying otherwise than for re-sale.

THE THIRD SCHEDULE, Part A, gives the list of feeding stuffs that may be mixed with those in Schedule I to produce compound cake, meal or mixture, *viz.*: meat and bone meal or meat meal or whale meat meal, molasses, home-grown wheat, barley, beans or peas (crushed or ground), and any other home-grown cereal or pulse (crushed or ground).

Part B gives the feeding stuffs that may be mixed with those in Schedule I to form concentrates: dried blood, meat meal, meat and bone meal, liver meal, whale meat meal, ground-nut cake meal, soya bean cake meal, wheat germ, cod-liver oil, any other vitamin potent oil, dried milk (whole, separated or buttermilk), dried whey, dried grass meal, dried clover meal, dried yeast, kelp.

THE FOURTH SCHEDULE gives a list of 17 centres in England and Wales with the distances from them beyond which mills are classified as "country flour mills."

* H.M. Stationery Office, York House, Kingsway, London, W.C.2. 1940. Price 5d. net.

STATUTORY RULES AND ORDERS. 1940. No. 1119*

The Compound and Mixed Feeding Stuffs (Control) (No. 2) Order, 1940.**Dated June 29, 1940**

THE Order contains 11 Articles and two Schedules. Article 1 defines the terms used in the Order:

"Compound or mixed feeding stuff" means any compound cake, compound meal, compound mixture or concentrate, or any other mixture of feeding stuffs.

"Feeding stuff" means any substance, other than hay or straw, which is used as food for livestock.

"Licensed concentrate" means a concentrate which a person is authorised to manufacture or prepare for sale in accordance with the terms of a licence granted under the Order.

"Livestock" means horses, mules, asses, cattle, sheep, pigs, goats, chickens, fowls, ducks, turkeys, guinea fowls, geese and carrier pigeons.

"Low protein oil seed cake or meal" means an oilseed cake or meal containing less than 22 per cent. of albuminoid.

"Protein rich substance" means a substance containing not less than 40 per cent. of albuminoid.

Article 2 orders that, except in accordance with the Minister's licence, no person shall:

- (a) Manufacture or prepare for sale during each three calendar months beginning July 1st, 1940, more than two-thirds of the total quantity of compound or mixed feeding stuff which he manufactured or prepared during the corresponding period of three months between July 1st, 1938, and June 30th, 1939.
- (b) Manufacture any compound or mixed feeding stuff other than one mentioned or in accordance with the conditions set out in the First Schedule, Part A or B.
- (c) Use in the manufacture or preparation of any compound or mixed feeding stuff described in Part A of the Schedule any ingredient other than one specified in the Second Schedule.

Article 3 orders that any licence granted under Article 2 may prescribe the composition and quality of any compound or mixed feeding stuff, the name or description under which it may be sold and its maximum price.

Article 4 allows feeding stuffs which are the property of the keeper of livestock to be mixed under specified conditions, subject to the keeper making a written request.

Article 5 provides that every person selling a compound or mixed feeding stuff other than a mixture mentioned in Article 4, shall, if the weight is 2 cwt. or more, cause to be affixed to each container thereof or furnished to the buyer on an invoice a statement specifying the description of the compound or mixed feeding stuff as set out in the first column of Part A or B of the First Schedule or as set out in a licence granted under Article 2 (b), and the name of the person who manufactured or prepared the compound or mixed feeding stuff.

Article 6 provides that nothing in this Order relieves any person from the duty of furnishing a statutory statement in accordance with Sec. 1 of the Fertilisers and Feeding Stuffs Act, 1926.

Article 8 revokes:

- (1) The General Licence dated September 3rd, 1939, as amended (S.R. & O., 1939, Nos. 1038 and 1777), made under the Control of Mills (Flour and Provender) No. 1 Order, 1939 (S.R. & O., 1939, No. 1037) so far as it is inconsistent with this Order.
- (2) The Compound and Mixed Feeding Stuffs (Control) Order, 1940 (S.R. & O., 1940, No. 864), but without prejudice to any proceedings in respect of any contravention thereof.

By Article 9 the Order does not apply to Northern Ireland.

Article 10 provides that Article 2 (a) of the Order shall come into force on July 1st, 1940, and that the whole Order shall come into force on July 15th, 1940.

THE FIRST SCHEDULE, PART A.—COMPOUND CAKES AND MEALS.—Briefly summarised, the particulars, tabulated in four columns, are as follows:

National Cattle Food, No. 1 (Dairy ration or cattle or sheep fattening).—Oil, min. 4, max. 6; albuminoids, min. 19.5, max. 21; fibre, max. 9.5 per cent.

National Cattle Food, No. 2 (Ration for grass feeding or fattening).—Oil, min. 4, max. 6; albuminoids, min. 14, max. 17; fibre, max. 12 per cent.

National Cattle Food, No. 3 (Rearing).—Oil, min. 5, max. 6; albuminoids, min. 20, max. 22; fibre, max. 8 per cent.

National Calf Gruel.—Oil, min. 6, max. 12; albuminoids, min. 15, max. 24; fibre, max. 7 per cent.

None of the foregoing compounds must contain more than 2 per cent. of lime (as CaCO_3) or 1 per cent. of salt (as NaCl).

National Pig Food, No. 1 (Pig-nuts or pig meal for sows or weaners).—Oil, min. —, max. 3·5; albuminoids, min. 16, max. 18; fibre, max. 7 per cent.

Composition: Maize meal and/or barley meal and/or dried potato products, min. 35, max. —; wheat feed (other than bran), min. 20, max. —; wheat, min. —, max. 20; oats, min. —, max. 10; bran, min. —, max. 10; low protein oilseed cake and/or meal, min. —, max. 20; *fish and/or animal protein rich substances, min. 5, max. —; molasses, min. —, max. 5; *sundries, including vitamin potent substances and mineral matter, min. —, max. 10 per cent. (salt not to exceed 0·5 per cent.); plus sufficient vegetable protein to give the required albuminoid content.

National Pig Food, No. 2 (Pig nuts or pig meal for fattening).—Oil, min. —, max. 4; albuminoids, min. 13, max. 15; fibre, max. 7 per cent.

Composition: Maize meal and/or barley meal and/or dried potato products, min. 35, max. —; wheat feed, min. 25, max. —; ground oats and/or bran, min. —, max. 20; wheat, min. —, max. 20; low protein oilseed cake and/or meals, min. —, max. 25; *fish and/or animal protein rich substances, min. 2·5, max. —; molasses, min. —, max. 5; *sundries (salt not to exceed 0·5 per cent.), min. —, max. 10 per cent.; plus sufficient vegetable protein to give the required albuminoid content.

National Poultry Food, No. 1 (*Summer Laying*).—Oil, min. 3, max. —; albuminoids, min. 17, max. 19; fibre, max. 8 per cent.

Composition: Wheat feed and/or low protein oilseed cake and/or meal, min. 40, max. 65; cereals, min. 25, max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *sundries (salt not to exceed 0·5 per cent.), min. —, max. 7·5 per cent.; plus sufficient vegetable protein to give the required albuminoid content. No decorticated or undecorticated cottonseed cake or meal may be used in this or any of the other poultry foods.

National Poultry Food, No. 1A (*Winter Laying*).—Oil, min. 3, max. —; albuminoids, min. 17, max. 19; fibre, max. 8 per cent.

Composition: Wheat feed and/or low protein oilseed cake and/or meal, min. 40, max. 65; cereals, min. 25, max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *cod-liver oil containing not less than 85 I.U. of vitamin D; *sundries including vitamin potent substances and mineral matter (salt not to exceed 0·5 per cent.), max. 7·5 per cent.; plus sufficient vegetable protein to give the required albuminoid content.

National Poultry Food, No. 2 (*Summer Growing*).—Oil, min. 3, max. —; albuminoids, min. 14, max. 17; fibre, max. 8 per cent.

Composition: Wheat feed and/or low protein oilseed cake and/or meal, min. 40, max. 65; cereals, min. 25, max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), max. 5; *sundries (salt not to exceed 0·5 per cent.), max. 7·5 per cent.; plus sufficient vegetable protein to give the required albuminoid content.

National Poultry Food, No. 2A (*Winter Growing*).—Oil, min. 5; max. —; albuminoids, min. 14, max. 17; fibre, max. 8 per cent.

Composition: Wheat feed and/or low protein oilseed cake and/or meal, min. 40, max. 65; cereals, min. 25, max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *cod-liver oil containing not less than 85 I.U. of vitamin D; *sundries (max. salt 0·5 per cent.), min. —, max. 7·5 per cent.; plus sufficient vegetable protein to give the required albuminoid content.

National Poultry Food, No. 3 (*Battery*).—Oil, min. 3, max. —; albuminoids, min. 15, max. 18; fibre, max. 8 per cent.

Composition: Wheat feed and/or low protein oilseed cake and/or meal, min. 35, max. 65; cereals, min. 30, max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *cod-liver oil containing not less than 85 I.U. of vitamin D; *sundries (max. salt 0·5 per cent.), min. —, max. 15 per cent.; plus sufficient vegetable protein to give the required albuminoid content.

National Baby Chick Food.—Oil, min. 3, max. —; albuminoids, min. 16, max. 18; fibre, max. 6·5 per cent.

Composition: Wheat feed and/or low protein oilseed cake and/or meal, min. 35, max. 65; cereals, min. 30, max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *cod-liver oil containing not less than 85 I.U. of vitamin D; *sundries (max. salt 0·5 per cent.), min. —, max. 20 per cent.; plus sufficient vegetable protein to give the required albuminoid content. Rye or rye products may not be used.

* These ingredients may be in the form of licensed concentrates. Other substances with as high vitamin D content may be used in place of cod-liver oil. Such substances must be warranted in writing as fully effective for poultry in accordance with the Chick Test of the British Standards Institution.

PART B—CEREAL MIXTURES.

National Cereal Mixture, No. 1.—Barley meal, min. 15; fine wheat feed and/or middlings and/or pollards, max. 25 per cent.; maize meal, sufficient to complete mixture.

National Cereal Mixture, No. 2.—Cattle ground oats, min. 30; fine wheat feed and/or middlings and/or pollards, max. 15 per cent.; maize meal, sufficient to complete mixture.

National Wheat Provender Mixture, No. 1.—Barley meal and/or maize meal, min. 65; wheat meal, max. 35 per cent.

National Wheat Provender Mixture, No. 2.—Fine wheat feed and/or middlings and/or pollards, max. 30; maize meal, min. 35; barley meal, min. 20 per cent.; wheat meal, sufficient to complete mixture.

National Cooked and Flaked Wheat Mixture.—Cooked and flaked maize, min. 75; cooked and flaked wheat, sufficient to complete mixture.

National Cereal Mixture, No. 3.—Maize meal and barley meal; no conditions as to percentages.

National Poultry Corn, No. 1A.—Wheat, max. 30; oats, min. 20 per cent.; cut and/or kibbled maize, sufficient to complete mixture.

National Poultry Corn, No. 1B.—Wheat, max. 30; clipped oats, min. 20 per cent.; cut and/or kibbled maize, sufficient to complete mixture.

National Poultry Corn, No. 2A.—Wheat, max. 30; oats, min. 20 per cent.; whole maize, sufficient to complete mixture.

National Poultry Corn, No. 2B.—Wheat, max. 30; clipped oats, min. 20 per cent.; whole maize, sufficient to complete mixture.

National Chick Feed, No. 1.—Fine cut wheat, max. 45; cut groats, min. 10 per cent.; No. 4 fine-screened maize grits, sufficient to complete mixture.

National Chick Feed, No. 2.—Cut wheat, max. 30; No. 3 fine screened maize grits, min. 30; whole groats, min. 10 per cent.; dari and/or millet, sufficient to complete mixture.

National Chick Feed, No. 3.—Fine cut wheat, max. 50 per cent.; No. 4 fine-screened maize grits, sufficient to complete mixture.

National Chick Feed, No. 4.—Cut wheat, max. 60; whole groats, min. 10 per cent.; No. 3 fine-screened maize grits, sufficient to complete mixture.

THE SECOND SCHEDULE.—No ingredient other than those in the following list and those contained in the First Schedule to the Feeding Stuffs (Maximum Prices) Order, 1940, as amended, may be used in the preparation of compound or mixed feeding stuffs (see Article 2 (c) p. 505):—Sugar cane molasses, dried grass meal, dried clover meal, alfalfa meal, lucerne meal, dried liver meal, feeding dried blood, dried yeast, cod-liver oil or other vitamin potent substances, licensed concentrates, buck-wheat, home-grown wheat, dredge corn, home-grown barley, home-grown peas, malt, biscuit meal, mineral matter, spices, herbs, liquorice root, dried milk, dried buttermilk, whey powder, whey paste, groats, ground tapioca root (including tapioca flour, manioc meal, mandioca meal, cassava meal and tapioca ampas of feeding quality), kapok seed cake, kapok cake meal, dari, millet, rye. All the cereals mentioned may be whole, crushed or ground.

STATUTORY RULES AND ORDERS. 1940. No. 1238***Order Dated July 11, 1940, Amending the Sausages (Maximum Prices) Order, 1940**

THE following is a Summary of this Order made by the Minister of Food:

1. The Sausages (Maximum Prices) Order, 1940* (hereinafter referred to as the "Principal Order") is amended as follows:

(a) The definition of "Pork Sausages" in Article 1 of the Principal Order is replaced by the following definition:

"Pork Sausages" means sausages of which at least 90 per cent. of the meat content consists of pork.

(b) After the definition of "Kosher beef sausages" in Article 1 of the Principal Order the following definitions are inserted:

"Pork sausage meat" means sausage meat of which at least 90 per cent. of the meat content consists of pork.

"Beef sausage meat" means sausage meat which is ordinarily known and sold as beef sausage meat, and includes sausage meat the meat of which is not beef alone.

"Kosher sausage meat" means sausage meat manufactured as respects the meat therein from beef obtained from cattle slaughtered in accordance with the Jewish practice of slaughter.

* H.M. Stationery Office, 1940. Price 1d. net.

* S.R. & O., 1939, No. 394 (cf. ANALYST, 1940, 65, 358).

- (c) At the end of Article 1 of the Principal Order the following paragraph is inserted:
 "Any sausages other than pork sausage and Kosher beef sausages as defined by this Order shall be deemed to be beef sausages for the purposes of the Order, and any sausage meat other than pork sausage meat and Kosher sausage meat as defined by this Order shall be deemed to be beef sausage meat for the purposes of the Order."
- (d) This allows a sum not exceeding 1d. per lb. to be added to the price of sausages (except Kosher beef sausages) filled in sheep casings and sold at not less than ten to the pound weight.
- (e) In paragraph 2 of Article 3 the words "or any Order made by the Minister" is substituted for the word "Order."
- (f) The word "or" is substituted for "of" in the last line of paragraph (5) of Article 5 in the Principal Order.
- (g) The following Articles are inserted in the Principal Order immediately after Article 5.
- 5A. Where it appears to the Minister of Food that an offence has been committed in respect of which proceedings might be taken under the Defence (General) Regulations, 1939, against some person for an infringement of this Order and the Minister is satisfied that the offence of which complaint is made was due to an act or default of some other person and that the first mentioned person could establish a defence under Article 5 of this Order, the Minister may cause proceedings to be taken against that other person without first causing proceedings to be taken against the first mentioned person.
- 5B. This Order does not apply to cooked or canned sausages.
2. The Principal Order as thus amended took effect on July 15th, 1940.

Analysis of Commercial Fats and Oils

REPORT OF AMERICAN CHEMICAL SOCIETY COMMITTEE*

THE Committee on Analysis of Commercial Fats and Oils recommends the adoption by the American Chemical Society of its report on the following six methods:

TITRE.—Vertical stirring is recommended as more convenient, and giving a sharper end-point than horizontal stirring. A new (partial immersion) thermometer covering a wider range is specified. It has been found that a differential temperature of 10° C. between the titre-point and the bath is insufficient for low titres, and it is recommended that this be increased. The proposed modifications do not give results differing from those obtainable by the horizontal stirring method.

DETECTION OF TRISTEARIN IN LARD (BÖMER NUMBER).—A modification of the official A.O.A.C. method for the detection of beef fat in lard is described. The A.O.A.C. capillary tube method for melting-points yields consistent and satisfactory results with glycerides and fatty acids, and is therefore recommended for this determination in place of the use of a sulphuric acid bath.

With care, sufficient crystals of definite composition can be obtained by crystallisation from acetone at 30° C. In the modified method described, 20 g. of the filtered sample are treated in a centrifuge tube or cylinder with acetone at 30° C., and the solution is made up to 100 ml. with acetone, shaken and left for 18 hours at 30° ± 2° C. The tube is then centrifuged for 5 minutes, or if a cylinder is used the supernatant liquid is siphoned off. Another 20-ml. portion of acetone (at 30° C.) is added to the crystals and the mixture is centrifuged (or the liquid siphoned off) as before. The washing with 20 ml. of acetone is repeated, but this time the mixture is transferred to a filter-paper, and the crystals are washed with 5 small portions of acetone at 30° C. After removal of as much acetone as possible by means of a vacuum pump the paper is removed from the funnel and, with its contents spread out, allowed to dry thoroughly at a temperature below the m.p. of the glycerides. The mass is then finely divided and its m.p. determined in a sealed capillary tube.

The remainder of the glycerides is saponified by boiling for an hour with 100 ml. of 0.05 N alcoholic potassium hydroxide solution in a 500-ml. Erlenmeyer flask with a small funnel in its neck. The soap solution is treated with 100 ml. of water, evaporated on the steam-bath to remove alcohol, transferred to a 500-ml. separating funnel, diluted to about 250 ml., neutralised with hydrochloric acid (1 + 1), a slight excess of which is added, and shaken with 75 ml. of ethyl ether. The aqueous layer is drawn off, and the ethereal layer is washed until neutral to methyl orange and evaporated on the steam bath, and the fatty acids are dried for a few minutes at 100° C.

For determining the m.p., the capillary tube is dipped into the melted acids so that the sample stands about 1 cm. high in the tube. The open end is then sealed in a gas flame, and the tube is left for 30 minutes in ice water or overnight in a refrigerator (4° to 10° C.). The tubes containing the fatty acids and the glycerides are attached to the thermometer by a rubber band, and

* *Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 379-384.

the thermometer is suspended in a beaker of water (suitably agitated), so that the bottom of the thermometer bulb is about 3 cm. below the level of the water, the temperature of which must be at least 10° C. below the m.p. of the sample. The water is heated to give a temperature increase of about 0.5° C. per minute; the m.p. is the point at which the samples become clear and liquid.

If the m.p. of the glycerides, plus twice the difference between the m.p. of the glycerides and that of the fatty acids, is less than 73° C. the lard is regarded as adulterated. The Committee's investigation has indicated that 10 per cent. of beef fat can be detected with certainty, and that amounts down to 5 per cent. may be found. The method is not applicable to hydrogenated pork fats.

SMOKE, FLASH AND FIRE POINTS.—It is recommended that the latest revision of methods of the American Society for Testing Materials be adopted. *Smoke Point.*—The sample is heated to within about 75° F. of the smoke point (the temperature at which a thin bluish smoke is continuously given off) in a Cleveland open flash cup (A.S.T.M. designation D92-33), after which the flame is regulated so that the temperature of the sample rises at a rate not less than 9° or more than 11° F. per minute. The flash cup fits into a circular opening in an asbestos board resting on a ventilated metal heating plate of specified dimensions in which there is also a central circular opening. The source of heat is centred under the opening in the plate and must be of a type not to produce local over-heating, and a cabinet of specified dimensions surrounds the heating device. An A.S.T.M. open flash thermometer is used.

FLASH AND FIRE POINTS.—The same apparatus, including the thermometer, is used, but without the cabinet. Exact details of the procedure are given.

VILLAVECCHIA TEST.—The following modified (A.O.A.C.) test is recommended:—Ten ml. of the sample are mixed with an equal volume of hydrochloric acid (sp.gr. 1.19) and shaken for 15 seconds with 0.1 ml. of the Villavecchia reagent (2 ml. of furfural in 100 ml. of 95 per cent. ethyl alcohol). If any colour is observed in the lower layer which separates, 10 ml. of water are added, and the mixture is again shaken. If the colour persists the test is positive; if it disappears sesame oil is not present.

MODIFIED GARDNER BREAK TEST FOR SOYA BEAN OIL.—Only two minor changes in the method are recommended: The use of a tall beaker helps to prevent loss by frothing, and the thermometer (A.S.T.M. Open Flash) recommended will ensure better control of the temperature.

HYDROXYL VALUE.—It is recommended that the method for calculating the value should be included in the description of the A.O.A.C. method.

COLOUR READING.—The Committee has approved the use of the 2.5-cm. (1-inch) column in the Lovibond system of colour reading for samples of oils and fats which cannot be read in a 13.335-cm. (5.25-inch) column.

International Union of Chemistry

RULES FOR NAMING INORGANIC COMPOUNDS*

A. GENERAL.—*Names and Formulae.*—When there is no risk of uncertainty, formulae should be used for complicated compounds. As a rule, indications of stoichiometric proportions are not necessary when systematic names are used. Trivial names (*e.g.* saltpetre, caustic soda) which are not incorrect are permissible, but incorrectly formed names (*e.g.* sulphate of magnesia) should not be used even in technical literature.

B. BINARY COMPOUNDS.—*I. Position of Constituents.*—The electropositive element, *e.g.* in salts and salt-like compounds should come first (in English and German names). The electro-negative constituent ends in "*ide*."

II. Indications of the Proportions of Constituents.—Stock's method of indicating valency (Roman figures in brackets) is advocated; *e.g.* "Copper (I) Chloride" for CuCl; "Copper (II) chloride" for CuCl₂. The terminations "*ous*" and "*ic*" should be entirely discarded. Valency of elements should be indicated by Roman figure just above the symbol on the right hand side (*e.g.* Cu^I salts; Fe^{III} compounds). Greek numerical prefixes without hyphens are used to indicate stoichiometric composition ("*mono*" can usually be omitted). "*Octa*" should be used for 8 and "*Ennea*" for 9. Greek prefixes above 12 are replaced by Arabic figures (without hyphens). Arabic figures are also used for fractions, although $\frac{1}{2}$ may be expressed by "*hemi*." The functional system may be used instead of the stoichiometric (*e.g.* nitrous anhydride). The formulae are preferable to awkward names, *e.g.* Na₁₃Hg₁₃ rather than "12 sodium 13 mercuride."

III. Designation of Intermetallic Compounds.—Formulae alone should be used and, if possible, should give the exact number of atoms. If the composition is variable a stroke should be placed over the formula; *e.g.* AuZn. Compounds whose composition is not constant are termed "non-Daltonian" compounds, to distinguish them from "Daltonian" compounds of constant composition.

* Report of the Committee (W. P. Jorissen, *Chairman*, H. Bassett, A. Damiens, F. Fichter, and H. Remy) for the Reform of Inorganic Nomenclature, June 3rd, 1940. 28, Rue St. Dominique, Paris.

IV. *Indicating the Mass, Atomic Number and State of Ionisation on the Atomic Symbols.*—The number of atoms is shown in the right-hand lower index; the state of ionisation in the right-hand upper index; the atomic number in the left-hand lower index; the mass in the left-hand upper index. Example:



V. *Group Names.*—Compounds of the halogens are to be termed *halogenides* (not haloids or halides). The elements oxygen, sulphur, selenium and tellurium may be termed *chalcogens* and their compounds *chalcogenides*.

The alkali metals should not be termed "alkalis" nor the alkaline earth metals "alkaline earths," but these terms are permissible abbreviations in compounds, *e.g.* alkali chloride.

C. *TERNARY, QUATERNARY, ETC., COMPOUNDS.*—The preceding rules for binary compounds apply also to compounds of more than two elements. Radicals with special names are treated like the elementary constituents of a compound. For example, $\text{Fe}(\text{SCN})_3$ = Iron (III) thiocyanate. Sulphur replacing oxygen in an acid radical should always be indicated by "thio" (not "sulpho"), but such names as "lead chlorofluoride" and "lead sulphochloride" are permissible for mixed salts.

The terms "alumino-," "boro-," "beryllo-," etc., silicates should be applied only to silicates which contain Al, B, Be, etc., in place of Si.

D. *OXYACIDS.*—Well-established names for most of the important simple oxyacids have been in use for a long time, and their alteration is unnecessary. The following names should be used for the more important acids of S, N, P and B:

H_2SO_3 = Sulphoxylic acid; $\text{H}_2\text{S}_2\text{O}_4$ = dithionous acid; H_2SO_2 = sulphurous acid; $\text{H}_2\text{S}_2\text{O}_3$ = thiosulphurous acid; $\text{H}_2\text{S}_2\text{O}_5$ = pyrosulphurous acid; H_2SO_4 = sulphuric acid; $\text{H}_2\text{S}_2\text{O}_6$ = thiosulphuric acid; $\text{H}_4\text{S}_2\text{O}_7$ = pyrosulphuric acid; H_2SO_5 = peroxy(mono)sulphuric acid; $\text{H}_2\text{S}_2\text{O}_8$ = dithionic acid; $\text{H}_2\text{S}_x\text{O}_6$ = polythionic acids; $\text{H}_2\text{S}_2\text{O}_8$ = peroxydisulphuric acid.

$\text{H}_2\text{N}_2\text{O}_2$ = hyponitrous acid; H_2NO_2 = nitroxylic acid; HNO_3 = nitrous acid; HNO_3 = nitric acid; HNO_4 = peroxyntic acid.

H_3PO_2 = hypophosphorous acid; H_3PO_3 = phosphorous acid; $\text{H}_4\text{P}_2\text{O}_5$ = pyrophosphorous acid; $\text{H}_4\text{P}_2\text{O}_6$ = hypophosphoric acid; H_3PO_4 = (ortho)phosphoric acid; $\text{H}_4\text{P}_2\text{O}_7$ = pyrophosphoric acid; HPO_3 = metaphosphoric acid; H_3PO_5 = peroxy(mono)phosphoric acid; $\text{H}_4\text{P}_2\text{O}_8$ = peroxydiphosphoric acid.

H_3BO_2 = borous acid; $\text{H}_3\text{B}_2\text{O}_4$ = hypoboric acid; H_3BO_3 = orthoboric acid; HBO_3 = metaboric acid; $\text{H}_2\text{B}_4\text{O}_7$ = tetraboric acid.

In notes on decisions in these lists it is pointed out that there was no justification for the use of the name "hydrosulphurous" acid for the compound $\text{H}_2\text{S}_2\text{O}_4$ after it had been shown that the salts have the composition $\text{R}_2\text{S}_2\text{O}_4$ and not RHS_2O_4 .

The name "hyposulphurous acid" would be justified for H_2SO_2 , but the well-established name "sulphoxylic acid" should be used. The names "hyposulphurous acid" and "hyposulphite" should be discarded.

A distinction must be made between acids or salts derived by substitution from hydrogen peroxide and those derived from the highest oxidation stages of some elements; the latter are correctly termed *per*-salts, whilst the former are named peroxy acids and peroxy salts.

The acid H_2NO_3 has been termed "hydronitrous acid," but is more correctly called "nitroxylic acid" by analogy with sulphonylic acid.

The term "ortho" is usually applied to the most hydroxylated acid known either in the free state or as salts or esters.

Pyro- and meta-acids are derived from ortho-acids by removal of water in stages. Pyro-acids have lost 1 mol. of H_2O from 2 mols. of ortho-acid (*e.g.* $\text{H}_2\text{S}_2\text{O}_5$, $\text{H}_2\text{S}_2\text{O}_6$, $\text{H}_4\text{P}_2\text{O}_5$, $\text{H}_4\text{P}_2\text{O}_6$). The rule does not apply to the polyboric acid $\text{H}_2\text{B}_4\text{O}_7$ ($= 2\text{B}_2\text{O}_3 \cdot \text{H}_2\text{O}$), which contains less water than metaboric acid HBO_3 ($= \text{B}_2\text{O}_3 \cdot \text{H}_2\text{O}$). To avoid breaking the rule, the acid $\text{H}_2\text{B}_4\text{O}_7$ should be termed tetraboric acid in agreement with the terminology suggested for isopoly acids.

Acids derived from oxy acids by replacement of O atoms by S atoms are to be termed thio-acids and their salts thio-salts; *e.g.* H_2CS_3 = trithiocarbonic acid.

When the hydrogen ion is considered to occur in the form $[\text{H}_3\text{O}]^+$, it is advisable to call it the *hydronium* ion (not *hydroxonium* ion).

E. *SALTS.*—I. *General.*—The name of the metal or electropositive radical should always precede that of the acid radical which terminates in -ate, -ite, or -ide. Names such as nitrate of silver are permissible, but the name of the metal oxide must not be used in place of that of the metal. For mixed or double salts the rules given under C apply.

Salts of nitrogen compounds.—If these are regarded as co-ordination compounds they are to be designated as -onium or -inium compounds; *e.g.* hydrazinium chloride. If, however, they are regarded as addition compounds, the rules derived for these will apply (see F. V).

II. *Acid Salts.*—The rational names are formed by using "hydrogen" for the H atoms present. The hydrogen is to be named last of the electropositive constituents. Formulae should be used for salts of complicated composition.

The term *acid salts* (monacid, diacid, etc.) may be used to emphasise the type of compound rather than its composition; also the expressions primary, secondary, tertiary, etc., salts.

III. *Basic Salts*.—If salts can be considered as addition compounds of hydroxides to neutral salts they are termed *hydroxy salts* (e.g. $\text{Cd}(\text{OH})\text{Cl}$ = cadmium hydroxychloride). But when the hydroxyl group is bound in a complex, Werner's nomenclature of *hydroxo-* or *ol-*groups should be used. Salts in which oxygen atoms, as well as acid radicals, are attached to the metal are termed *oxy salts*, although the names of radicals with special designations may also be used (e.g. BiOCl = bismuth oxychloride or bismuthyl chloride).

F. HIGHER ORDER COMPOUNDS.—I. *Complex (Co-ordination) Compounds*.—Stock's method of indicating valency has been extended to the Werner nomenclature. For *complex cations* the Roman figures for valency are placed in brackets after the name of the element to which they refer; e.g. $[\text{Cr}(\text{OH})_6]\text{Cl}_3$ = hexaquo chromium (III) chloride. For complex anions of salts or acids the valency of the central atom is given in brackets after the name of the complex which ends in -ate; e.g. $\text{H}_4[\text{Fe}(\text{CN})_6]$ = hydrogen hexacyanoferrate (II). It is unnecessary to give the valency of the central atoms of neutral compounds (non-electrolytes). Atoms or groups co-ordinated in the complex are to be mentioned in the following order: (i) *acidic groups*, such as cyano (CN), cyanato (NCO), oxalato (C_2O_4) and hydroxo (OH); (ii) *neutral groups*: aquo (H_2O), substituted amines [$\text{C}_2\text{H}_5(\text{NH}_2)_2$ = en], and last of all ammine (NH_3).

II. *Isopolyacids and their Salts*.—It is proposed that the empirical formulae should always be resolved into the Base anhydride:Acid anhydride ratios, thus avoiding unsettled questions of constitution. Alternatively, the composition referred to the simplest empirical formula, is given by means of Greek numerical prefixes, as in B II. The following examples are illustrative:

NaB_3O_6 Sodium pentaborate $\text{Na}_2\text{O} \cdot 5\text{B}_2\text{O}_3$ Sodium (1:5) borate
 $\text{Na}_4\text{Si}_2\text{O}_{10}$ Tetrasodium trisilicate $4\text{Na}_2\text{O} \cdot 3\text{SiO}_2$ Sodium (4:3) silicate
 $\text{Na}_{10}\text{Mo}_2\text{O}_{41}$ Decasodium dodecamolybdate $5\text{Na}_2\text{O} \cdot 12\text{MoO}_3$ Sodium (5:12) molybdate
 $\text{Na}_2\text{W}_2\text{O}_7$ Disodium ditungstate $\text{Na}_2\text{O} \cdot 2\text{WO}_3$ Sodium (1:2) tungstate
 NaVO_3 Sodium (mono)vanadate $\text{Na}_2\text{O}_5 \cdot \text{V}_2\text{O}_5$ Sodium (1:1) vanadate.

III. *Heteropolyacids and their Salts*.—The formulae are resolved into their constituent base and acid anhydrides, the simplest formula which expresses the analytical composition being used. Examples are:

$2\text{R}_2\text{P}_2\text{O}_5 \cdot 17\text{WO}_3$ = 17-Tungstodiphosphate or $5\text{R}_2\text{O} \cdot \text{P}_2\text{O}_5 \cdot 17\text{WO}_3$ = 17-Tungsto-2-phosphate.

$\text{R}_8\text{Si}_2\text{O}_5 \cdot 12\text{WO}_3$ = Dodecatungstosilicate or $4\text{R}_2\text{O} \cdot \text{SiO}_2 \cdot 12\text{WO}_3$ = 12-Tungstosilicate

IV. *Double Salts*.—The order of cationic constituents should be that of decreasing electro-positive character. Constituents common to both salts should be mentioned only once.

V. *Hydrates, Ammoniates and other Addition Compounds*.—The collective names: hydrate, peroxyhydrate (not perhydrate) and ammoniate should be applied to compounds containing molecules of H_2O , H_2O_2 or NH_3 . If the molecule forms part of a complex, the compounds are to be named *aquo*, *peroxyhydrate* and *ammine compounds* (see F. I). Addition compounds containing PCl_5 , NOCl , H_2S , $\text{C}_2\text{H}_5\text{OH}$, etc., are better shown by a formula than by a special name.

Fruit and Vegetable Preservation Research Station, Campden

ANNUAL REPORT FOR 1939

THE Director (Mr. F. Hirst, M.Sc.) in his introduction to the Report points out that since the beginning of the war the National Mark Scheme has been allowed to lapse, and that unless that scheme is revived or some alternative method of control is introduced there will be no standards for quality in canned fruits and vegetables. It would be to the ultimate good of the industry that there should be some means of quality control.

Investigations, described in detail in the Report, include the following:

DRAINED WEIGHT OF ENGLISH CANNED FRUITS AND VEGETABLES.—In continuation of previous work, *Annual Report*, 1938, p. 17; ANALYST, 1939, 64, 596) the Director and W. B. Adam give results showing the effect of the size of the mesh of the draining sieve on the drained weight. With a sieve with 8 openings to the inch the weights (expressed as percentage of filled weight) were on the average greater by 0.5 per cent. than those obtained with a mesh with 15 openings to the inch. For the sake of uniformity the suggested standards might well be based on the larger mesh.

The National Mark filling weights for A2 cans are 55 to 56 per cent. of the aqueous capacity of the cans for currants and berry fruits (except gooseberries), 59 per cent. for gooseberries, and 60 per cent. for stone fruits. The experiments described show that the filling weights of most soft fruits could be increased. It is suggested that for gooseberries, strawberries, raspberries, loganberries and blackberries the filling weight could be 59 per cent. of the capacity of the can, for blackcurrants, 58 per cent., and for cherries, plums and damsons, 61 per cent. The new standards, however, should be based, not on these filling weights, but on the final drained weights.

The following average drained weights (expressed as percentages of the aqueous capacity of the cans) were (i) observed and (ii) calculated in tests on 6000 cans of different sizes packed under the National Mark in recent years:—Gooseberries, (i) 56, 57, 55; (ii) 53, 56, 57. Strawberries, (i) 39, 37, 36; (ii) 34, 35, 36. Raspberries, (i) 46, 44, 45; (ii) 42, 44, 45. Loganberries, (i) 46, 45; (ii) 43, 45. Blackcurrants, (i) 52, 49; (ii) 46, 49. Blackberries, (i) 47, 44; (ii) 43, 45. Sweet cherries, (i) 52, 49; (ii) 51, 54. Greengages, (i) 56; (ii) 56. Golden plums, (i) 55, 51, 51; (ii) 49, 52, 53. Purple plums, (i) 55, 49; (ii) 50, 51. Victoria plums, (i) 55, 49; (ii) 52, 53. Damsons, (i) 54, 54; (ii) 54, 55.

The tests on vegetables were less comprehensive than those for canned fruits, each of the statistics being calculated from only 20 to 70 cans. The following average (i) observed and (ii) calculated drainage figures are recorded:

Beans (dwarf), whole, (i) 53, 52; (ii) 55, 54; sliced, (i) 56, 56; (ii) 59, 60. Beetroot, whole, (i) 66, 68; (ii) 67, 69; sliced, (i) 67, 71; (ii) 66, 67. Broad beans, ungraded, (i) 65; (ii) 69. Carrots, whole, (i) 60, 62; (ii) 62, 63. Celery, hearts, (i) 57, 63; (ii) 62, 65. Peas, (i) 61, 62, 63; (ii) 64, 65, 65. Potatoes, whole, (i) 63, 67; (ii) 67, 70.

The problem of standardising methods of determining drained weights has recently been studied by the U.S. Food and Drug Administration (Bonney, *J. Assoc. Off. Agric. Chem.*, 1939, 32, 370), and it is advisable also to explore the possibility of establishing minimum standards for English canned fruits and vegetables.

STUDY OF HYDROGEN SWELLS.—Recent work has shown that corrosion in lacquered cans occurs chiefly in the seams, where the metal is exposed. Adjustment of the minor constituents of the steel (*e.g.* sulphur, phosphorus, copper) to their optimum proportions might effect improvements. Experiments indicated that the time taken for high-copper low-phosphorus steels to form 10 per cent. of hydrogen swells would be roughly double that of steels of the present composition. The results of Hoar, Morris and Adam (*J. Iron and Steel Inst.*, 1939, 140, 55*r*) suggest that steel with a copper-content of 0.16 to 0.22 per cent. and a phosphorus-content below 0.04 per cent. is the most suitable for tinplate for lacquered cans for fruit, and it ought to be possible for manufacturers to supply such tinplate.

Sulphur compounds, present as ultramarine in "blued" sugar or as sulphite, act as accelerators of corrosion. Un-blued sugar should therefore always be used for canning. It is not known definitely at what concentration of sulphur dioxide corrosion is appreciably hastened, but possibly 10 to 20 p.p.m. in the sugar might have this effect.

The use of the post-lacquered (or flush lacquered) can affords almost complete protection against hydrogen swells for about 3 years under normal storage conditions and virtually eliminates the unsightly blue discoloration of red or purple fruits.

FRUIT GUMMING OF VICTORIA PLUMS.—The results of the tests (by the Director and W. B. Adam) showed no significant differences between plums grown on light or heavy soils. On the whole there was less gum when the crops were heavy and the fruit small. The percentage of surface gum fell as the fruits ripened, but early ripening fruits showed more gum than under-ripe fruits. Gumming was distributed unevenly over the trees and differed appreciably on neighbouring trees.

BEHAVIOUR OF TRACES OF SULPHUR IN CANNED FRUITS.—In the experiments (by G. Horner) the development of sulphide in presence of sulphite was followed by distilling the acidified fruit in an atmosphere of carbon dioxide, collecting the hydrogen sulphide in neutral silver nitrate solution, and titrating this with standard alkali. In lacquered cans the amount of sulphide present after 5 to 6 months did not exceed a trace, but in plain cans amounts up to 0.8 p.p.m. were recorded. The difference in the behaviour of the two types of can may be due to the unequal rates at which the changes from sulphur to hydrogen sulphide and from the latter to metallic sulphide take place.

HYDROGEN ION CONCENTRATION OF CANNED VEGETABLES.—Further progress in this investigation by W. B. Adam is recorded. In all the results described the quinhydrone electrode was used in combination with a saturated KCl-calomel half-cell, the E.M.F. being measured by means of a Tinsley potentiometer connected with a null-point galvanometer. The reading was taken about 30 seconds after adding the quinhydrone.

The preliminary results show that the pH value of immature peas does not alter appreciably during the later stages of ripening, although smaller sizes tend to have lower values than the larger grades. The pH of harvested peas is not affected by the stage of maturity at which they are cut. There is a fall in the pH of the covering liquid during processing, but with peas and beans the value subsequently rises as buffering substances are extracted. The following averages and ranges of final pH values for the principal English canned vegetables are given; the numbers of cans examined ranged from 10 to 82.

Beans, broad, 5.90–6.13 (av. 6.01); beans, dwarf, 5.33–5.99 (av. 5.59); beetroot, 4.94–5.61 (av. 5.39); carrots, 4.88–5.46 (av. 5.12); celery, 5.15–5.62 (av. 5.36); peas, immature, 5.67–6.37 (av. 6.12); potatoes, new, 5.68–6.24 (av. 5.82); spinach, 5.43–6.25 (av. 5.83); macedoine, 5.16–5.76 (av. 5.34).

HEAT PENETRATION IN ROTATING CANS.—To accelerate the passage of heat towards the centre of cans methods of mechanical agitation are used, including automatic reel-and-spiral pressure cookers and continuously rotating batch sterilisers. A rotating thermo-couple has been designed for measuring the rate of heat penetration in all sizes of cans, and is described and

illustrated in a communication by W. B. Adam and J. Stanworth. It was found that heat penetration in canned beans in tomato sauce was very slow. Thus in stationary cans it required 300 minutes to reach a central temperature of 235° F. with a processing temperature of 240° F., but when the cans were rotated at 5 rev. per 10 seconds, with 20 seconds stop, the same central temperature was reached in 12½ minutes. The results indicated that with A10 cans the safe processing time may be reduced from 3½ hours to half-an-hour by rotating the cans.

The new apparatus may also be used in testing other products to control the destruction of the spores of *Byssoschlamys fulva*.

Bibliography on Metals in Foods and Biological Materials

(Supplementing the series published in the ANALYST up to 1933, 58, 340, and bringing the Bibliography up to date.)

I. COBALT

- AHMAD and McCOLLUM. Cobalt Content of some Food Materials from different parts of the United States. *Amer. J. Hyg.*, 1939, 29, (A), 25; Abst., ANALYST, 1939, 64, 287. (Modification of Stare and Elvehem method; results given for peas, beans and cereals.)
- ANON. Cobalt Deficiency in Sheep and Cattle. *New Zealand J. Agr.*, 1939, 59, 585; Abst., C.A., 1940, 34, 4431. (Cobalt preparations for stock and soil described.)
- ASKEW. Cobalt Deficiencies at Nelson, New Zealand. *New Zealand J. Sci. Tech.*, 1939, 20, 302A. Successful Use of Cobalt Salts for Pasture Top-dressing at Glenhope, *id.*, 315A; Value of Cobalt Supplements for Breeding Ewes at Sherry River, Nelson, *id.*, 192A; Absts., B.C.A., 1940, 49AIII, 75B, 83B.
- ASKEW and DIXON. Influence of Cobalt Top-dressing on the Cobalt Status of Pasture Plants. *New Zealand J. Sci. Tech.*, 1937, 18, 688; Abst., C.A., 1937, 31, 6388. (Cobalt content of plants increased by cobalt chloride treatment of soil; large amounts toxic to plants.)
- ASKEW and DIXON. Value of Cobalt Salts for Pasture Top-dressing in the Treatment of Stock Ailment at Glenhope, Nelson and Morton Mains, Southwold. *New Zealand J. Sci. Tech.*, 1937, 19, 317; Abst., B.C.A., 1938, 831b.
- ASKEW and MAUNSELL. Cobalt Content of some Nelson Pastures. *New Zealand J. Sci. Tech.*, 1937, 19, 337; Abst., C.A., 1938, 32, 5133.
- ASTON. Micro-methods for Cobalt and Zinc. *New Zealand Dept. Agr., Ann. Rept. Chem. Section*, 1935-36; Abst., ANALYST, 1937, 62, 43. (Cobalt by nitroso-R-salt, to 0.001 mg.; iron separated.)
- ASTON. Zinc and Cobalt in Animal Specimens. *Id.*; Abst., ANALYST, 1937, 62, 42. (Cobalt in sheep livers; bush-sick about 0.06, healthy 0.20 p.p.m.)
- ASTON. Zinc and Cobalt in Pasture Samples. *Id.*; Abst., ANALYST, 1937, 62, 42. (Not detected in several samples from sick and healthy areas.)
- BABICKA. Colorimetric Determination of Cobalt in Nutrient Solutions and Plant Ashes. *Mem. Soc. roy. sci. Bohême*, 1935, 1934, 1; *Ber. ges. Physiol. exptl. Pharmacol.*, 88, 401; Abst., C.A., 1937, 31, 7920. (Colorimetric with ammonium thiocyanate; hydrochloric acid solution of ash neutralised with lithium hydroxide.)
- BECKER and GADDUM. Composition of Limonite effective and ineffective in correcting "Bush-sickness" in Cattle. *J. Dairy Sci.*, 1937, 20, 737; Abst., C.A., 1938, 32, 1304. (Cobalt required.)
- BELL. Some Physiological Aspects of the Cobalt Problem. *New Zealand J. Sci. Tech.*, 1937, 18, 716; Abst., C.A., 1937, 31, 6297.
- BERTRAND and NAKAMURA. Investigation of the Physiological Importance of Nickel and Cobalt. *Bull. Soc. sci. hyg. aliment.*, 1936, 24, 338; Abst., C.A., 1937, 31, 4385.
- BERTRAND and NAKAMURA. Physiological Importance of Nickel and Cobalt. *Ann. Inst. Pasteur*, 1934, 53, 371; Abst., B.C.A., 1934, 1395A.
- BROWNING. Manganese and Cobalt in Plant and Animal Economy. *Sci. Progress*, 1937, 31, 276; Abst., B.C.A., 1937, 441AIII. (Survey.)
- CHASE. Detection of Heavy Metals and Traces of Impurities in Textile Fabrics. *Amer. Dyestuffs Rept.*, 1933, 22, 75; Abst., C.A., 1933, 27, 2584. (Microscopic or spot tests in acid solution; cobalt mentioned.)
- CLAYTON. Detection of Metals in Fibrous Materials, Dyes and Organic Pigments. *J. Soc. Dyers & Colorists*, 1937, 53, 380; Absts., B.C.A., 1937, 1327B; C.A., 1938, 32, 360. (Precipitation [with nickel] as sulphide; potassium xanthate test.)
- CORNER and SMITH. Effect of Cobalt on Pine Disease in Sheep. *Biochem. J.*, 1938, 32, 1800; Abst., B.C.A., 1938, 1023AIII.
- DIXON. Use of Cobaltised Salt Lick in the Control of a Lamb Ailment at Morton Mains, Southwold. *New Zealand J. Sci. Tech.*, 1937, 18, 892; Abst., C.A., 1937, 31, 8013.

- DUVAL and LE GOFF. Occurrence of Small Amounts of Cobalt in Human Urine. *Compt. rend.*, 1937, **204**, 817; Abst., *B.C.A.*, 1937, 202AIII. (Cobalt above 0.05×10^{-6} g.; Precipitated with α -nitroso- β -naphthol in acetic acid; complex salts first electrolysed.)
- FILMER and UNDERWOOD. Enzootic Marasmus. *Australian Vet. J.*, 1937, **13**, 57; Abst., *C.A.*, 1937, **31**, 6703. (Cobalt as cure.)
- GODDEN. Methods for the Chemical Analysis of Biological Material in Nutritional Investigations. *Imp. Bur. Animal Nutrition*, 1937, *Tech. Commun.* No. 9; Abst., *C.A.*, 1938, **32**, 4199. (Contains compilation of methods for elements in foods; cobalt mentioned.)
- GODDEN. "Trace" Elements in Human and Animal Nutrition. *Chem. and Ind.*, 1939, 791; Abst., *B.C.A.*, 1940, 231AIII. (Physiological properties; cobalt mentioned.)
- GRIMMETT. Cobalt in Animal Organs. *New Zealand Dept. Agr., Ann. Rept. Chem. Section*, 1938-9; Abst., *ANALYST*, 1940, **65**, 34. (Sheep livers: bush-sick about 0.04 or less, healthy above 0.10 p.p.m.) Cobalt Deficiency in Pastures. *Id.*; Abst., *ANALYST*, 1940, **65**, 33. (Cobalt minimum in late spring and early summer; maximum in winter or after long drought.) Standards for Trace Elements in Pastures. *Id.*; Abst., *ANALYST*, 1940, **65**, 34. (Chemical and spectrographic analyses; cobalt mentioned.)
- GRIMMETT. Cobalt in Foodstuffs. *New Zealand Dept. Agr., Ann. Rept. Chem. Section*, 1936-7; Abst., *ANALYST*, 1938, **63**, 113. (Determinations made; some results in abst.)
- GRIMMETT. Deficiency Diseases of Livestock: Bush Sickness. *New Zealand Dept. Agr., Ann. Rept. Chem. Section*, 1937-8; Abst., *ANALYST*, 1939, **64**, 38. (Pasture analysis, McNaught's method: Bush-sick cattle and sheep, 0.01 to 0.04, sheep 0.04 to 0.07, healthy above 0.07 p.p.m. cobalt.)
- HARVEY. Cobalt Status of some W. Australian Soils and Pastures. *J. Dept. Agr., W. Australia*, 1937, **14**, 386; Abst., *C.A.*, 1938, **32**, 7201. (Pastures: Healthy 0.03 to 0.30, sick regions less than 0.08, p.p.m. of cobalt.)
- HELLER, KUHLE and MACHEK. Determination of Traces of Heavy Metals in Mineral Waters. *Mikrochem.*, 1937, **23**, 78; Abst., *ANALYST*, 1938, **63**, 374. (Polarographic; cobalt mentioned.)
- HOPKIRK. Cobalt-feeding Experiment at Arohana. *New Zealand J. Agr.*, 1937, **54**, 344; Abst., *C.A.*, 1937, **31**, 7531.
- HOPKIRK. Deficiencies known and suspected in Livestock Nutrition of New Zealand. *New Zealand J. Agr.*, 1936, **53**, 200; Abst., *B.C.A.*, 1938, 586b.
- HOPKIRK. Importance of Cobalt. Relation to Health of Farm Animals. *New Zealand Dept. Agr.*, 1938, **56**, 21; Abst., *B.C.A.*, 1938, 710b. (Review.)
- KIDSON and ASKEW. Critical Examination of the Nitroso-R-salt Method for the Determination of Cobalt in Pastures. *New Zealand J. Sci. Tech.*, 1940, **21b**, 178; Abst., *C.A.*, 1940, **34**, 4689. (Reliable: details [given] very important.)
- KIDSON, ASKEW and DIXON. Colorimetric Determination of Cobalt in Soils and Animal Organs. *New Zealand J. Sci. Tech.*, 1936, **18**, 601; Abst., *B.C.A.*, 1937, 706b. (Modification of nitroso-R-salt method.)
- KIDSON and MAUNSELL. Effect of Cobalt Compounds on the Cobalt Contents of Supplementary Fodder Crops. *New Zealand J. Sci. Tech.*, 1939, **21a**, 125; Abst., *C.A.*, 1940, **34**, 568; *B.C.A.*, 1940, 306b.
- KONISHI and TSUGE. Mineral Matters of certain Leguminous Crops. I. Inorganic Constituents of Underground Parts of certain Leguminous Crops. *Mem. Coll. Agr., Kyoto Imp. Univ.*, No. 37 (Chem. Ser. No. 20), 1936, 1; Abst., *C.A.*, 1936, **30**, 4541. (Traces detected spectroscopically; cobalt mentioned.)
- LINES. Effect of Ingestion of Minute Quantities of Cobalt by Sheep affected with "Coast Disease." *J. Coun. Sci. Ind. Res., Australia*, 1935, **8**, 117; Abst., *B.C.A.*, 1935, 1008a.
- LUGG and JOSLAND. Colorimetric Determination of Cobalt in Solution by the Nitroso-R-Salt. *Australian J. Exp. Biol. Med. Sci.*, 1936, **14**, 319; Abst., *B.C.A.*, 1937, 150A1. (Stare and Elvehjem method modified in presence of calcium chloride.)
- MCNAUGHT. Cobalt Content of Limonites used in the Treatment of Bush Sickness. *New Zealand Dept. Sci. Ind. Res., Chem. Section, Dept. Agr.*, 1937; cf. *ANALYST*, 1937, **62**, 467. (Modified Stare and Elvehjem method after removal of iron; detects down to 0.02 mg. of cobalt.)
- MCNAUGHT. Cobalt Content of N. Island Pastures. *New Zealand J. Sci. Tech.*, 1938, **20**, 14a; Abst., *C.A.*, 1938, **32**, 8494. (Down to 0.05 γ determined in pastures; formula for cobalt complex with nitroso-R-salt revised.)
- MCNAUGHT. Determination of Cobalt in Animal Tissues. *ANALYST*, 1939, **65**, 23. (Acid digestion; modified nitroso-R-salt determination.)
- MCNAUGHT and PAUL. Seasonal Variations in Cobalt Content of N. Island Pastures. *New Zealand J. Sci. Tech.*, 1939, **21**, 95b; Abst., *B.C.A.*, 1940, 387b. (Generally higher in closely grazed pasture than in mature grass.)
- MACPHERSON and STEWART. Photometric Determination of Cobalt. *Biochem. J.*, 1938, **32**, 763; Abst., *B.C.A.*, 1938, 273A1. (As nitroso-R-salt complex.)
- MARSTON. Cobalt in Ruminant Nutrition. *Ann. Rev. Biochem.*, 1939, **8**, 570.
- MARSTON, LINES, THOMAS and McDONALD. Cobalt and Copper in Ruminant Nutrition. *Nature*, 1938, **141**, 398; Abst., *B.C.A.*, 1938, 412AIII.

- MARSTON, THOMAS, MURNAME, LINES, McDONALD, MOORE and BULL. Studies on Coast Diseases of Sheep in S. Australia. *Australian Counc. Sci. Ind. Res.*, 1938, *Bull.* No. 113; Abst., *B.C.A.*, 1938, 594AIII.
- MAZÉ. Rôle of Special Elements (Boron, Copper, Zinc, Manganese, etc.) in Plant Nutrition. *Ann. Rev. Biochem.*, 1936, 5, 525; Abst., *C.A.*, 1936, 30, 5615. (Cobalt mentioned.)
- NEAL and AHMANN. Cobalt as an Essential Element in Animal Nutrition. *Science*, 1937, 86, 225; Abst., *B.C.A.*, 1937, 472AIII.
- NEAL and AHMANN. Essential Nature of Cobalt in Bovine Nutrition. *J. Dairy Sci.*, 1937, 20, 741; Abst., *B.C.A.*, 1938, 212AIII.
- PATTERSON. Cobalt and Sheep Disease. *Nature*, 1937, 140, 363; Abst., *B.C.A.* 1937, 418AIII (0.20 p.p.m. cobalt found in sick pastures.)
- PATTERSON. Some Observations on a Disease of Sheep on Dartmoor. *Empire J. Exptl. Agr.*, 1938, 6, 262; Abst., *C.A.*, 1938, 32, 8493. (Cobalt deficiency.)
- RAMAGE. Analysis of Tissue for Metallic Content. *Nature*, 1936, 138, 762; Abst., *B.C.A.*, 1936, 1532A; *C.A.*, 1937, 31, 1061. (Extraction with N-hydrochloric acid before spectrographic determination; cobalt mentioned.)
- RAMAGE. Biological Distribution of Metals. *Nature*, 1936, 137, 67; Abst., *B.C.A.*, 1936, 258A, and *C.A.*, 1936, 30, 2605. (Spectroscopic determination in foods; cobalt detected in beans.)
- RICEMAN and McDONALD. Preliminary Investigations on the Effect of Copper and other Elements on the Growth of Plants in a "Coasty" Calcareous Sand at Robe, S. Australia. *Australian Counc. Sci. Ind. Res.*, 1938, Pamphlet 78; Abst., *B.C.A.*, 1938, 970AIII. (Cobalt does not benefit plants, but its application controls "coast disease" of livestock.)
- RIGG. Mineral Content of Pastures. Cobalt Investigations at the Cawthron Inst. *New Zealand Dept. Sci. Ind. Res., Ann. Repts.*, 1935-6 and 1938-9; Absts., *C.A.*, 1937, 31, 5024; 1940, 34, 1113.
- ROST. Concentrations of the Chemical Elements by Organisms. *Přiroda*, 1936, 29, 223; Abst., *C.A.*, 1939, 33, 653. (Concentrations in plant and animal tissues summarised; cobalt mentioned.)
- ROTHAMSTED EXP. STATION. Cobalt Deficiencies in Soils. *Chem. Dept. Rept.*, 1938; Abst., ANALYST, 1940, 65, 105. (Pasture deficiency and sheep disease.)
- SHELDON. Mineral Basis of Life. *Brit. Med. J.*, 1934, I, 47; Abst., *C.A.*, 1934, 28, 2035. (Review of physiological effects of metals; cobalt mentioned.)
- STANTON and KIDSON. Cobalt Status of Soils and Pastures in the Sherry and Wangapeka Districts, Nelson. *New Zealand J. Sci. Tech.*, 1939, 21, 65B; Abst., *B.C.A.*, 1940, 387B. (General, but not invariable, correlation of cobalt content of pastures and soils.)
- STEWART and PONSFORD. Pining in Sheep not Curable by Cobalt Administration. *Nature*, 1940, 145, 1023.
- SULLIVAN. Inorganic Constituents of Wheat and Flour. *Cereal Chem.*, 1933, 10, 503; Abst., *C.A.*, 1934, 28, 1412. (Cobalt in wheat 0.01 p.p.m.)
- SYLVESTER and LAMPITT. Determination of Cobalt in Foods. *J. Soc. Chem. Ind.*, 1940, 59, 57; Abst., ANALYST, 1940, 65, 424. (Cobalt determined colorimetrically by modified McNaught method; traces, —0.0002 mg.—detected by adsorption column. Results given.)
- UNDERWOOD. Significance of the "Trace" Elements in Nutrition. *Nutrition Abstracts and Reviews*, 1939-40, 9, 515. (Review. Spectroscopic studies; cobalt mentioned.)
- UNDERWOOD and FILMER. Determination of the Biologically Potent Element (Cobalt) in Limonite. *Australian Vet. J.*, 1935, 11, 84; Abst., *B.C.A.*, 1936, 1140A. (Enzoötic Marasmus caused by cobalt deficiency in animal foods.)
- UNDERWOOD and HARVEY. Enzoötic Marasmus. Cobalt Content of Soils, Pastures and Animal Organs. *Australian Vet. J.*, 1938, 14, 183; Abst., *C.A.*, 1939, 33, 3947.
- V. ZEPPELIN and GLASS. Cobalt as a Cure for Disease of Stock on Pasture. *Ernähr. Pflanze*, 1938, 34, 186; Abst., *C.A.*, 1938, 32, 6697. (Disease causes decreased milk production; cobalt as preventative.)
- WALL. Trace Elements in Relation to Bush Sickness. *New Zealand Dept. Sci. Ind. Res., Ann. Rept. Chem. Section Dept. Agr.*, 1935-6; Abst., ANALYST, 1937, 62, 466.
- WEBB. II. Spectrographic Analyses of Marine Invertebrates, with special reference to Chemical Composition of their Environment. *Sci. Proc. Roy. Dublin Soc.*, 1937, 21, 505.
- WEBB and FEARON. Studies on the Ultimate Composition of Biological Material. I. Aims, Scope and Methods. *Id.*, 21, 487. (Graphite electrodes cause error in spectrographic analysis; Absts., *C.A.*, 1937, 31, 6268. (Cobalt mentioned.)
- WUNSCH. Trace Elements in Livestock Disease. *Chem. and Ind.*, 1939, 531; Abst., *B.C.A.*, 1939, 765AIII. (Cobalt discussed.)
- WUNSCH. Tracking down a Deficiency Disease. *Chem. and Ind.*, 1937, 855; Abst., *C.A.*, 1938, 32, 628. (Cobalt deficiency.)

British Standards Institution

The following Standard Specification has been issued:*

No. 914—1940. BRITISH STANDARD TESTS FOR LABORATORY PORCELAIN.

The tests have been prepared by a committee representative of makers and users of laboratory porcelain appointed by the Technical Committee on Scientific Glassware and Laboratory Ware. Several of the tests are based on those adopted by the Sub-Committee of the Institute of Chemistry and published in 1920 (*J. and Proc. Inst. of Chem.*, 1920, [3], 210-214). A new rapid test of the resistance to heat and cooling has been introduced.

The tests are divided into five sections: (1) Appearance, shape, weight, etc. (2) Tests for porosity of body and imperfections in glaze (dye test). (3) Resistance to heat and sudden change of temperature. (4) Constancy of weight and resistance of glaze to high temperatures. (5) Resistance of glaze to acid and alkali.

The test for porosity consists in soaking the specimens, some whole, others broken, in a 0.5 per cent. aqueous solution of eosin for 18 hours and ascertaining the amount of staining, if any.

A special furnace is described and illustrated for testing the resistance of ordinary crucibles to heat. The crucible is placed bottom downwards in a cage suspended from the lid of the furnace and heated at the specified temperature for 15 minutes, after which it is allowed to drop into a bucket of water at 15° C., which is placed 6 inches below the end of the furnace. The crucible should not break or crack after being heated to 240° C. and suddenly cooled under the specified conditions.

In the high temperature tests clean broken pieces of the porcelain are gently ignited in crucibles of the same make, and the vessels are cooled, weighed, and heated for 2 hours in a muffle at 950°-1000° C. There should be no change in weight and no adhesion of the pieces to one another or to the crucible.

Resistance of the glaze to acid and alkali is determined by testing the dishes in a specified manner with hydrochloric acid of constant b.p. for 4 hours, with *N* sodium carbonate solution for 4 hours at 100° C., and finally with 5 per cent. caustic soda solution under the same conditions. The losses, expressed as mg. per sq. dcm., should not exceed 1 mg. for HCl, 10 mg. for Na₂CO₃ or 60 mg. for NaOH.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Colorimetric Determination of the Preservative Value of Hops. Standard Colour Values of Some Hybrid Hops. A. A. D. Comrie. (*J. Inst. Brewing*, 1940, **46**, 255-256.)—It is pointed out that in the author's colorimetric method for the determination of hop preservative values (*ANALYST*, 1939, **64**, 828) the mixed α - and β -resins present must be considered in terms of the " α -resin equivalent," i.e. the effective sum of the resins when the usually-accepted preservative value of 1/3 of that of the α -resin is allotted to the β -resin. Thus α and β per cent. of α - and β -resin, respectively, in a hop would correspond with an " α -resin equivalent" of $(\alpha + \beta/3)$ per cent. Similarly, the "standard colour value" (S.C.V.) of a hop is, by definition (*loc. cit.*), the colour value of 0.8 mg. of the α -resin equivalent as it exists in the hop. Since, however, it has been found that the S.C.V. for the α -resin from all sources hitherto examined is a constant, (95) under the conditions of the experiment, whilst the S.C.V. for the β -resin depends on the nature of the hop in question, this means that the S.C.V. for the mixture will also vary according to the nature of the hop. For

ordinary English varieties, however, this variation has been found to be negligible, and an average value of 82 for the α -resin equivalent $(\alpha + \beta/3)$, could be used. The new hybrid variety "Brewers' Gold" proved an exception (S.C.V., 95), and further determinations have therefore been made on some of the better-known hybrids raised at Wye College. Values found for the S.C.V. of the weight of hop containing 0.8 mg. of α -resin equivalent were, Brewers' Gold, 96; Bullion Hop, 95; Quality Hop, 94; Brewers' Favourite, 91; Early Promise, 93; and Fillpocket, 92. These high figures appear to be due to the high S.C.V. of the β -resins (see above), but this in turn does not result from the influence of the species *Humulus americanus* in the inter-specific hybrids, because the wild uncultivated *H. americanus* (var. *neomexicanus*) grown at Wye had an S.C.V. of only 82, whilst a further hybrid similar to those above had an S.C.V. of 84. It is suggested that if these hybrid hops come into commercial cultivation a S.C.V. of 94 should give results in fair accordance with the accepted formula for the gravimetric method, $10(\alpha + \beta/3)$; comparisons of the preservative values obtained by the gravimetric and colorimetric methods confirm this for the above 6 hops. J. G.

* To be obtained from the Publications Department, British Standards Institution, 28, Victoria Street, London, S.W.1. Price 2s. net, post free 2s. 3d.

Fatty Acids and Glycerides of Solid Seed Fats. IX. *Mimusops Heckelii* (Baku) Kernel Fat. D. Atherton and M. L. Meara. (*J. Soc. Chem. Ind.*, 1940, 59, 95-96).—*Mimusops Heckelii*, N.O. *Sapotaceae*, is a tree widely distributed in the closed forest areas of the Gold Coast. The nuts, of average weight 20 g., consisted of about 60 per cent. of hard pericarp enclosing 40 per cent. of kernel containing 52 per cent. of fat with the following constants:—saponification equiv. 295.4, iodine value, 52.7; acid value, 2.4; unsaponifiable matter, 0.4 per cent. The component fatty acids in the neutral fat were determined by distillation of the methyl esters; another portion of neutral fat was crystallised from acetone, and the three fractions of increasing solubility were examined. The proportions of fatty acids were computed to be: palmitic, 4.4; stearic, 36.0; arachidic, 0.5; hexadecenoic, 0.3; oleic, 58.5; linolic, 0.3 per cent. by weight. The component glycerides were approximately: steardiolein, 41-47; oleodistearin, 32-26; palmitodiolein, 14-6; triolein, 10-12; oleopalmitostearin, 2-8; fully saturated palmitodistearin 1 mol. per cent. From this analysis and those of other fats, e.g. shea butter, it is concluded that when oleic acid approaches 60 per cent. of the mixed fatty acids in a seed fat, triolein is usually present in proportions which, although relatively small, are larger than those of the fully saturated components when the converse relationship holds, i.e. the rule of even distribution is less closely followed in the production of the unsaturated glycerides than of the fully saturated glycerides. D. G. H.

Component Fatty Acids and Glycerides of some *Myristica* Fats. D. Atherton and M. L. Meara. (*J. Soc. Chem. Ind.*, 1939, 58, 353-357).—The kernel fats of (a) *Virola surinamensis* from S. America and (b) *Pycnanthus Kombo* (*Myristica angolensis* Walw.) from Sierra Leone were examined. The composition of the nuts was: shell, (a) 17, (b) 14; kernel, (a) 83, (b) 86 per cent., and the kernels yielded to petroleum spirit (a) 71 and (b) 61.6 per cent. of dark brown solid fats. These had saponification equiv. (a) 246.1, (b) 249.9; iodine value, (a) 14.5, (b) 67.0; free fatty acids as oleic acid, (a) 17.2, (b) 18.7; iodine value of neutral fat after removal of highly unsaturated resin acids, (a) 9.9, (b) 32.3. The component acids, estimated by ester fractionation, consisted of (per cent. wt.) decoic, (a) 0.5, (b) 0.2; lauric, (a) 14.8, (b) 11.5; myristic, (a) 72.5, (b) 58.1; palmitic, (a) 4.9, (b) 9.1; tetradecenoic, (a) —, (b) 26.3; oleic, (a) 6.3, (b) 17; unsaponifiable, resins, (a) 1.0, (b) 3.4. Crystallisation from acetone gave fractions the major component glycerides of which were estimated approximately as:—tetradecenodimyrystin, (a) —, (b) 33; trimyrystin, (a) 43, (b) 24; laurodimyrystin, (a) 31, (b) 17; oleolaurodimyrystin, (a) 12; lauromyristopalmitin, (a) 10, and in (b) small amounts of oleotetradecenomyristin, myristoditetradecenoic and laurotetradecenomyristin. The free fatty acids of both fats contained higher pro-

portions of unsaturated acids than those from the neutral glycerides. The kernel fat of *Pycnanthus Kombo* is at present unique in containing 24 per cent. of Δ^4 -tetradecenoic (myristoleic) acid, an acid not previously reported in seed fats but present in fish and whale oils, and to a small extent in butter-fat. The isomeric dihydroxy acids, m.p. 123° and 81.5° C. respectively, were prepared by mild alkali and acid oxidation of this tetradecenoic acid. D. G. H.

Fatty Acids and Glycerides of the Seed Fats of *Allanblackia floribunda* and *Allanblackia parviflora*. M. L. Meara and Y. A. H. Zaky. (*J. Soc. Chem. Ind.*, 1940, 59, 25-26).—*Allanblackia floribunda* (a) from Nigeria, and *Allanblackia parviflora* (b) from the Gold Coast belong to the N.O. *Guttiferae*. The nuts of *A. parviflora* consisted of 35.5 per cent. of shell and 64.5 per cent. of kernel containing 69.4 per cent. of fat. The analytical constants of the two fats were:—saponification equiv., (a) on refined fat 297.7, (b) 295.3; iodine value (a) on refined fat, 35.4, (b) 37.2; free fatty acids as oleic acid, (b) 1.9 per cent. The fats were examined in the usual way by lead salt separation from alcohol and fractionation of the methyl esters, and the proportions of fatty acids were calculated as:—myristic, (a) —, (b) 1.5; palmitic, (a) 2.9, (b) 2.3; stearic, (a) 57.1, (b) 52.0; arachidic, (a) 0.2, (b) 0.3; oleic, (a) 39.4, (b) 43.9; linolic, (a) 0.4, (b) —. Crystallisation from acetone and examination of the resulting fractions gave data for calculating the glyceride composition to be approximately:—oleodistearin, (a) 76, (b) 60; steardiolein, (a) 15.5, (b) 26-29; oleopalmitostearin, (a) 5.0, (b) 6-9 mol. per cent. Thus the rather small differences in the proportions of fatty acid components in the two fats give rise to greater differences in those of the major components of the glycerides. The fat of *A. parviflora* is seen to be very similar to that of *A. Stuhlmannii* (*J. Soc. Chem. Ind.*, 1931, 50, 468T; Abst., ANALYST, 1932, 57, 113). D. G. H.

Composition of Commercial Palm Oils. V. Partial Separation of Palm Oils by Crystallisation as an Aid to the Determination of the Component Glycerides. T. P. Hilditch and L. Maddison. (*J. Soc. Chem. Ind.*, 1940, 59, 67-71).—Two more palm oils have now been examined (cf. *J. Soc. Chem. Ind.*, 1935, 54, 77T; Abst., ANALYST, 1935, 60, 328). The two oils: (a) a plantation oil from the Cameroons and (b) a native oil from Grand Bassa, differed in their respective contents of palmitic and oleic acid, and after preliminary separation by acetone into glyceride fractions of relatively simple composition were found to contain the following component fatty acids: myristic, (a) 1.1, (b) 0.6; palmitic, (a) 45.1, (b) 37.6; stearic, (a) 4.1, (b) 3.7; hexadecenoic, (a) 0.8, (b) 1.4; oleic, (a) 38.6, (b) 50.3; linolic, (a) 10.3, (b) 6.4 per cent. by weight. The component glycerides were

approximately (mol. per cent.) "oleo" dipalmitin, (a) 43, (b) 31; palmitodi"olein," (a) 3, (b) 41; "oleo" palmitostearin, (a) 11, (b) 10; tri"olein," (a) 6, (b) 12; tripalmitin, (a) 5, (b) 3; dipalmitostearin, (a) 3, (b) 3; stearodi"olein," (a) perhaps up to 1, (b) perhaps traces. Comparison of these figures with those obtained before by progressive hydrogenation (*loc. cit.*) shows that the oils form a series in which the proportion of palmitic acid rises whilst that of oleic acid falls. Summarising, the chief components of palm oils are regarded as "oleo" dipalmitin and palmitodi"olein" in amounts varying according to the proportions of palmitic, oleic and linolic acids in the whole fats. These together usually amount to 70-75 per cent. of the palm oil, "oleo" dipalmitin preponderating in oils of high palmitic acid content and conversely. The minor components are "oleo" palmitostearin (about 10-15) linoleodi"olein" and/or tri"olein" (6-15, varying with the oleic + linolic acid content of the palm oil), and tripalmitin + dipalmitostearin (3-9 per cent.) varying with the palmitic acid content of the palm oil. D. G. H.

Fat of Land Crabs (Seychelles Islands). T. P. Hilditch and K. S. Murdi. (*J. Soc. Chem. Ind.*, 1939, 58, 351-353.)—The fat, extracted from land crabs in the Seychelles, consisted of 101.3 g. of a soft yellow solid with saponification equiv. 234.4, iodine value 19.1, free fatty acids, as oleic acid, 1.0, and unsaponifiable matter 0.3 per cent. The fat was systematically crystallised from stated volumes of acetone at 0° C. for 1 day, the mixed fatty acids from each fraction were converted into the methyl esters, and each group was distilled at 0.1 mm. pressure through an electrically heated and packed column. From the data obtained the component acids were computed to be: octoic, 1.5; deoic, 5.3; lauric, 47.5; myristic, 14.0; palmitic, 13.1; stearic, 1.7; tetradeconoic, 0.7; hexadecenoic, 2.2; oleic, 5.3; linolic, 1.5; unsaturated C_{20-22} acids, 2.2 mol. per cent. The fat contained 66.3 per cent. of fully saturated glycerides of which the component acids included octoic, 3.2; deoic, 7.1; lauric, 54.8; myristic, 20.5; palmitic, 12.7; stearic, 1.7 mol. per cent. The striking resemblance between the fatty acids and those of seed fats of the palm family, and the almost entire absence of the characteristics of a typical marine animal fat are probably accounted for by the fact that these crabs feed on coconuts. D. G. H.

Alkaloids of some *Chondrodendron* Species and the Origin of *Radix Pareirae Bravae*. H. King. (*J. Chem. Soc.*, 1940, 737-746.)—Differences in the optical rotatory power of bebeerine from different samples of the drug *Radix Pareirae Bravae* are attributed to its origin in two very similar species, *Chondrodendron platyphyllum* and *Ch. microphyllum*. These yield *l*- and *d*-bebeerine, respectively, the latter occurring in the drug on the English market. From these species,

from *Ch. candicans* (the British Guiana species) and from the drug, alkaloids were obtained as follows:—*Ch. platyphyllum* (Rio) root (924 g.):—*d*-iso-chondrodendrine (sulphate 43.1 g. plus base 1.7 g.), *l*-bebeerine (9.0 g.), *Ch. platyphyllum* (Bahia), root: (720 g.), *l*-bebeerine (hydrochloride 55.3 g. plus base 1.50 g.), *d*-isochondrodendrine (hydrochloride 1.1 g. plus base 0.65 g.); stems (538 g.): *l*-bebeerine (28.32 g.); leaves (300 g.): *l*-chondrofoline, *d*-isochondrodendrine, *l*-bebeerine (total crude alkaloid 3.4 g.) *Ch. microphyllum* (Bahia), root (845 g.): *d*-isochondrodendrine (sulphate 18.3 g. plus base 1.42 g.), *d*-bebeerine (hydrochloride 15.4 g. plus base 1.5 g.). *Ch. candicans* (British Guiana), stems (1500 g.): *d*-isochondrodendrine (hydrated sulphate 17.7 g. plus base 1.48 g.), *d*-bebeerine (4.93 g.). *Pareirae Bravae*: *d*-bebeerine *d*-isochondrodendrine, *d*-isococlaurine. The widely differing proportions of alkaloids in the same species are ascribed to climatic and seasonal causes. The new alkaloids *l*-chondrofoline and *d*-isococlaurine are related to bebeerine and isomeric with coclaurine respectively. Chondrofoline, $C_{23}H_{35}O_6N$, is phenolic and contains three methoxyl groups; methylation yields an amorphous *O*-methyl methiodide and methochloride closely resembling the corresponding products from bebeerine, while other reactions give products identical or enantiomorphic with those from bebeerine. The alkaloid does not give the Millon reaction and contains only one phenolic group. Isococlaurine, which was isolated from a relatively large amount of *Radix Pareirae Bravae*, is phenolic and contains one methoxyl group. The derivative on complete methylation, *d*-*O*-dimethyl-*N*-methylcoclaurine methiodide, is the optical enantiomorph of the similar product from natural *l*-coclaurine. The alkaloid gives the Millon reaction and does not give the catechol reaction. Probable structural formulae for the two alkaloids are given. The study of *d*-isochondrodendrine has enabled probable structures to be assigned to proto-curidine and neoprotocuridine, isomeric phenolic alkaloids of pot-curare; many bisbenzylisoquinoline alkaloids can also be classified as either bebeerine or isochondrine types.

E. B. D.

Separation and Determination of Isomeric Menthols. R. T. Hall, J. H. Holcomb, Jr., and D. B. Griffin. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 187-188.)—The synthetic menthol used to replace Menthol U.S.P. (*l*-menthol from *Mentha arvensis*) consists of varying mixtures of the other isomers with some of the U.S.P. product, the number and proportion of isomers depending upon the method of synthesis. When *d*-citronellal (from citronella oil) is the initial material a mixture of 3 isomers, *viz.* *d*-neomenthol, *d*-isomenthol and *l*-menthol, is obtained, and the large proportion of *l*-menthol in this particular mixture makes its commercial separation possible. The analytical reactions of these isomers have been studied. Most methods for the determination of total menthol depend

upon acetylation followed by hydrolysis of the acetylated product with alcoholic potassium hydroxide solution. Saponification with a solution of potassium hydroxide in diethylene glycol (Redemann and Lucas, *Ind. Eng. Chem., Anal. Ed.*, 1937, 9, 521; *Abst.*, ANALYST, 1938, 63, 62) reduces the time of hydrolysis to one-fourth or one-fifth. The rates of both the acetylation and the hydrolysis vary for the different isomers. Acetylation for 2 hours is sufficient, and low results obtained for some of the isomers were invariably due to incomplete hydrolysis. Hydrolysis of the esters of *l*-menthol and *d*-isomenthol with 0.5 *N* alcoholic potassium hydroxide was practically complete within 30 minutes, but the *d*-neomenthol ester required 3.5 hours. The more rapid hydrolysis effected by the diethylene glycol solution of potassium hydroxide did not affect the accuracy of the results; again, however, the *d*-neomenthol ester (and consequently the isomeric mixture) required a longer time for hydrolysis than the esters of the other isomers.

A. O. J.

Biochemical

Estimation of Phosphorus. R. J. L. Allen. (*Biochem. J.*, 1940, 34, 858-865).—The method of Fiske and Subbarow (*J. Biol. Chem.*, 1925, 66, 375; *cf.* ANALYST, 1926, 51, 205), as modified by King (*Biochem. J.*, 1932, 26, 292; *Abst.*, ANALYST, 1932, 57, 532), was not found quite satisfactory when the analysis depended on the measurement of absolute colour density. The following method, in which amidol is substituted for 1-amino-2-naphthol-4-sulphonic acid, is proposed instead. Not more than 20 ml. of the solution to be tested (containing about 0.4 mg. of phosphorus in the form of phosphate) are transferred to a 25-ml. volumetric flask, and 2 ml. of 60 per cent. perchloric acid are added, followed by 2 ml. of amidol reagent (2 g. of amidol and 40 g. of pure sodium bisulphite are dissolved in glass-distilled water and diluted to 200 ml.), and 1 ml. of 8.3 per cent. ammonium molybdate solution. The solution is made up to volume and, after standing for 5 to 30 minutes, its extinction coefficient is measured. The amount of phosphorus is calculated by reference to a standard curve prepared by similar treatment of standard phosphate solutions. To measure the amount of total phosphorus, an appropriate amount of the material is digested over a micro-burner in a micro-Kjeldahl flask with 2.2 ml. of perchloric acid. When the liquid is colourless (a few drops of hydrogen peroxide may be added to facilitate the oxidation), the flask is allowed to cool, and the contents are washed into a 25-ml. volumetric flask and treated with amidol and ammonium molybdate solutions as before. Some materials gave turbid or coloured solutions, and the following further treatment was then adopted. The blue solution, obtained by allowing the solution of phosphate to stand for 5 to 30 minutes with the perchloric

acid and amidol and ammonium molybdate solutions, is treated with 1 ml. of 10 per cent. oxalic acid solution. The mixture is transferred to a separating funnel and gently shaken with about 10 ml. of isobutyl alcohol. The aqueous phase is re-extracted with a further 5 ml. of isobutyl alcohol, the combined extracts are diluted to 25 ml. with ethyl alcohol, and the extinction coefficient of the solution is measured within 24 hours. The amount of phosphorus is calculated by the use of a standard curve.

F. A. R.

Quantitative Estimation of Glycuronic Acid and its Conjugated Compounds by means of the Naphthoresorcinol Test of Tollens. W. Mozolowski. (*Biochem. J.*, 1940, 34, 823-828).—Numerous workers have endeavoured to make Tollens' naphthoresorcinol test applicable to the quantitative estimation of glycuronic acid, but without success. The following method has been found to give satisfactory results:—A 0.2 per cent. aqueous solution of naphthoresorcinol is allowed to stand overnight and filtered; the reagent must be used within a day. The solution under examination, made up to 2 ml. if necessary, is mixed in a test-tube with 2 ml. of the naphthoresorcinol reagent and 2 ml. of conc. hydrochloric acid are added from a burette. The tube is heated for 30 minutes in a boiling water-bath and cooled in ice-water for 10 minutes. To each sample 2 ml. of ethyl alcohol are added, and the mixtures are transferred to 100-ml. glass-stoppered measuring cylinders, the test-tubes being washed out 3 times with 5 ml. of pure ether. The cylinders are shaken 10 times in 30 seconds, and the absorption of the ethereal layer is measured in a Pulfrich photometer, using a 2-cm. cell and filter S57; a control solution prepared by treating distilled water in the same way is used in the other cell. The concentration of glycuronic acid is only proportional to the extinction coefficient, however, within the range 0.0125 to 0.05 mg., so that a preliminary test must be made when the concentration is unknown. Samples of 2.0, 1.0 and 0.5 ml. are respectively made up to 2.0 ml. and treated as described above. If the amount of glycuronic acid present lies between 0.005 and 0.5 mg., the extinction values obtained will be proportional to the volumes taken. If such proportionality does not exist, the solution to be examined must be diluted 5 or 10-fold and the procedure repeated until proportionality is reached. In this way satisfactory results were obtained on pure solutions of glycuronic acid, glycurone, benzoylglycuronic and bornylglycuronic acids. The method can also be applied to blood and urine, but account must be taken of interfering substances, and in urines containing such substances the estimation can only be carried out if the concentration of glycuronic acid is sufficiently high to allow of adequate dilution. True values can, of course, only be obtained when there is proportionality within a given series.

F. A. R.

Blood Sugar Levels in Rats receiving the Cataractogenic Sugars Galactose and Xylose. W. J. Darby and P. L. Day. (*J. Biol. Chem.*, 1940, **133**, 503-509.)—A modification of the method devised by Folin and Malmros (*J. Biol. Chem.*, 1929, **83**, 115) for the estimation of blood sugar is described; this can be carried out on 0.025-ml. samples of blood. Blood from the tail-vein of the experimental animals was drawn to the 0.025 ml. mark of a special diluting pipette, and the solution was diluted to the 2.5 ml. mark with dilute tungstic acid solution (20 ml. of 10 per cent. sodium tungstate solution are diluted to 800 ml. and mixed with 20 ml. of 2/3 *N* sulphuric acid, and the solution is diluted to 1 litre). After being mixed, the solution was expelled into a clean dry 15-ml. centrifuge tube and centrifuged. Two ml. of the supernatant liquid were transferred to a test-tube graduated at 12.5 ml., 2 ml. of a standard solution containing 0.01 mg. of glucose per ml., were introduced into a similar tube, and to each were added 1 ml. of 0.4 per cent. potassium ferricyanide solution and 0.5 ml. of a carbonate-cyanide solution (prepared by mixing 150 ml. of 1 per cent. sodium cyanide solution with a solution of 8 g. of anhydrous sodium carbonate and diluting to 500 ml.). The solutions were heated in boiling water for 8 minutes, cooled for 2 minutes, and then treated with 2.5 ml. of colloidal ferric iron solution (prepared by allowing 20 g. of soluble gum ghatti, supported by a wire screen, to soak overnight in 1 litre of water, and adding to the extract a solution of 5 g. of anhydrous ferric sulphate in 75 ml. of 85 per cent. phosphoric acid diluted with 100 ml. of water. About 15 ml. of 1 per cent. potassium permanganate solution are added to destroy certain reducing materials present in gum ghatti). Finally, the contents of each tube were diluted to the 12.5 ml. mark and mixed, and the colours were compared in a colorimeter with a yellow filter. F. A. R.

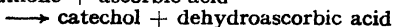
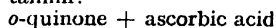
Glutamic Acid of Normal and Malignant Tissue Proteins. A. C. Chibnall, M. W. Rees, E. F. Williams and E. Boyland. (*Biochem. J.*, 1940, **34**, 285-300.)—Kögl and Erxleben (*Z. physiol. Chem.*, 1939, **258**, 57) claimed that malignant tissues contain a number of their constituent amino-acids in a partially racemised form, whereas normal tissues contain amino-acids of normal rotation. The difference was said to be particularly striking in respect of the glutamic acid present. Thus acid hydrolysis of normal lung or heart tissue yielded *l*(+) glutamic acid, showing the normal rotation (in 9 per cent. hydrochloric acid) of $[\alpha]_D + 31.6^\circ$, whilst hydrolysis of various kinds of tumours gave rotations as low as $+4.6^\circ$, indicating the presence of as much as 40 per cent. of *d*(-) glutamic acid. The method used by Kögl and Erxleben for isolating glutamic acid as its hydrochloride was the cuprous oxide method of Abderhalden and Fuchs (*Z. physiol. Chem.*, 1908, **57**, 339), which has now been shown to give comparatively small yields of glutamic acid with pref-

erential salting out of racemic glutamic acid hydrochloride. Thus even from normal tissue protein preparations, such as gliadin, small first crops of partly racemised glutamic acid hydrochloride were obtained. Glutamic acid was isolated from seven different malignant tissue protein preparations and two normal tissue protein preparations by a modification of Foreman's method (*Biochem. J.*, 1914, **8**, 463). Good yields of the acid were obtained, and the degree of racemisation was in every instance small, the amount of the *d*(-) antipode from the malignant tissue being only about 0.2 per cent. It is concluded that Kögl and Erxleben's claims cannot be sustained and that racemisation of amino-acids is not characteristic of the proteins of malignant tissue. The modified Foreman method used in the present investigation was as follows: The dry protein was hydrolysed with 20 per cent. hydrochloric acid for 22 hours, and the excess of hydrochloric acid was removed by repeated evaporation *in vacuo*. The resulting syrup was treated with cream of lime, the excess of lime was removed, and the calcium salts were precipitated from the solution by the addition of 8.5 volumes of absolute alcohol. The calcium salts were dissolved in water, re-precipitated with alcohol and re-dissolved in water. Calcium was removed from the solution by means of oxalic acid, and the filtrate was evaporated to a syrup after addition of hydrochloric acid. On standing overnight much glutamic acid hydrochloride crystallised out, and was recrystallised from water. The mother liquors were boiled with sulphate-free copper carbonate, the excess was removed by filtration, and the filtrate was concentrated. Impure copper aspartate separated and was purified by re-dissolving in hydrochloric acid and re-precipitating with copper carbonate. The filtrate from the copper aspartate was freed from copper by means of hydrogen sulphide, and the calcium salts of the remaining amino-acids were isolated and treated as before, yielding a further crop of glutamic acid hydrochloride. This procedure was applied to all mother liquors, and ultimately nearly the whole of the glutamic acid was isolated. F. A. R.

Sucrase Activity in the Barley Plant. H. K. Archbold. (*Biochem. J.*, 1940, **34**, 749-763.)—Component parts of the barley plant were collected from the time of emergence of the sixth leaf until harvest, frozen in solid carbon dioxide, and stored in the frozen condition until required. The sucrase content of each portion was estimated by powdering the frozen material, allowing it to warm up to room temperature and squeezing out the sap. Aliquot portions of the green suspension thus obtained were incubated with a sucrose solution in presence of an acetate buffer solution; after neutralising with sodium carbonate solution, the optical rotation of the solution was measured in a polarimeter, and the amount of sucrose hydrolysed was calculated. The optimal pH for barley sucrase was found to be

4.8. The lowest level of activity was found in the stems, a slightly greater activity in the roots, whilst the leaves and leaf-sheaths were about twice as active as the stems. In the ear, activity increased rapidly with growth and declined again after full emergence. The maximum value for the ear was higher than for any other part of the plant. The total activity of the plant increased until ear emergence and then decreased. F. A. R.

Fermentation Process in Tea Manufacture. V. Cytochrome Oxidase and its Probable Role. VI. Effect of Dilution on the Rate and Extent of Oxidations in Fermenting Tea Leaf Suspensions. E. A. H. Roberts. (*Biochem. J.*, 1940, **34**, 500-516.)—V. Subsequent work has revealed that the mechanism previously proposed (*cf. ANALYST*, 1939, **64**, 616) for the fermentation process of tea is untenable. This hypothesis postulated that the oxidation of ascorbic acid precedes that of tannin during fermentation. It has now been shown that tea leaf at the end of fermentation still retains a considerable part of its enzymatic activity, taking up oxygen if fresh substrate, in the form of a green leaf infusion, is added; ascorbic acid, on the other hand, is oxidised very slowly by fermented tea leaf, the rate of oxygen uptake being much lower than that recorded at the beginning of fermentation. Moreover, the failure to absorb oxygen is not due to inhibition by tannin fermentation products. It would appear, therefore, that the oxidation of ascorbic acid by fermenting tea leaf is a side-reaction brought about by the primary oxidation product of tea tannin:



and that ascorbic acid oxidase is not responsible for the first stage of fermentation. Previously the author had found it impossible to reconcile his results (obtained in Assam) and his hypothesis with the results obtained by Lamb (*Tea Res. Inst. Ceylon, Ann. Rep.*, 1937, 65), working in Ceylon. By repeating the experiments at a lower temperature, comparable with the temperature prevailing in the hills in Ceylon, Lamb's results have been confirmed. The new results provide conclusive proof that an oxidase system occurs in tea leaf and is readily inactivated by alcohol at temperatures of about 30° C., and furnish additional evidence against the ascorbic acid theory. The properties of the oxidase now shown to be present are in close agreement with those of cytochrome oxidase. Thus (1) both enzymes are readily destroyed by alcohol and acetone, (2) both are firmly attached to tissues, (3) the oxygen uptake of each is markedly accelerated by an increase in the oxygen tension and (4) neither is very specific (unlike ascorbic acid oxidase and catechol oxidase); both oxidise catechols, tea tannin, *p*-phenylenediamine and ascorbic acid. On the assumption that cytochrome oxidase is the enzyme responsible for the uptake of oxygen, it becomes necessary to modify the scheme formerly proposed,

substituting cytochrome *c* (Fe''- and Fe'''-haematin denote the reduced and oxidised forms respectively and CoH₂ and Co denote the reduced and oxidised forms of cozymase) for ascorbic acid as the oxygen carrier:

- (1) $\text{Fe}''\text{-haematin} + \text{O}_2 \xrightarrow{\text{cytochrome oxidase}} \text{Fe}'''\text{-haematin} + \text{H}_2\text{O}_2$
- (2) $\text{Tea tannin} + \text{Fe}'''\text{-haematin} \rightarrow o\text{-quinone} + \text{Fe}''\text{-haematin}$
- (3) $\text{Tea tannin} + \text{H}_2\text{O}_2 \xrightarrow{\text{peroxidase}} o\text{-quinone}$
- (4) $o\text{-Quinone} \rightarrow \text{condensation products}$
- (5) $\text{Fe}'''\text{-haematin} + \text{CoH}_2 \rightarrow \text{Fe}''\text{-haematin} + \text{Co}$
- (6) $o\text{-Quinone} + \text{CoH}_2 \rightarrow \text{tea tannin} + \text{Co}$
- (7) $\text{Co} + \text{substrates} \xrightarrow{\text{dehydrogenases}} \text{CoH}_2 + \text{reaction products}$

VI. The rate of oxygen uptake by suspensions of fermenting tea leaf in water increases as the dilution increases; if the logarithm of the concentration is plotted against the logarithm of the oxygen uptake, a straight line results. From this graph it is possible to calculate the oxygen uptake for any given concentration, such as that existing on the fermenting room floor under factory conditions. The agreement between the calculated and the observed values was found to be fairly close. The variation in the rate of uptake with enzyme concentration is not due to diffusion effects, since tea juice itself shows a similar behaviour; nor is it due to variations in the concentration of substrate. The most likely explanation appears to be that it is due to variations in the degree of dispersion of the coenzymes required for carbohydrate breakdown. This view is supported by the observation that where coenzyme dilution can have no effect, as in the simple oxidation of tea tannin or pyrogallol by the cytochrome system, the reaction is independent of enzyme concentration, but direct evidence for this explanation is as yet lacking. The fermentation of tea in the factory does not follow the same course as fermentation in dilute aqueous suspension, peroxidase playing a negligible role in dilute suspension. Thus the essential reactions in the factory are (3), (5) and (6) above, whilst in dilute suspension the essential reactions are (2) and (6). F. A. R.

Vitamin A, Carotene and Xanthophyll Content of the Yolk of Hens' Eggs. B. Sjöllema and W. F. Donath. (*Biochem. J.*, 1940, **34**, 736-748.)—Different groups of hens were given the same basal diet with different sources of carotene and vitamin A. The amounts of vitamin A, carotene and xanthophyll in the eggs laid by these hens were estimated after saponification of the yolks and extraction with peroxide-free ether. Half of the extract from each yolk was evaporated to

dryness, the residue was dissolved in chloroform, and the solution was treated with antimony trichloride solution; the total blue colour due to the three components was measured in a Pulfrich step-photometer. The other half of the extract was evaporated to dryness, the residue was dissolved in 75 ml. of petroleum spirit (b.p. 60° to 80° C.) and the solution was shaken several times with 85 per cent. alcohol until no further colour was extracted. The petroleum spirit solution, containing the carotene, was washed with water, dried and distilled. The residue was dissolved in a suitable volume of petroleum spirit, and the colour of the solution was measured in the step-photometer; filter S47 being used. The combined alcoholic extracts, containing the xanthophyll, were diluted with 5 volumes of water and extracted with petroleum spirit. The extract was washed, dried and concentrated to a suitable volume, and the colour of the solution was measured as before, with the use of filter S47. The carotene and xanthophyll contents were estimated by reference to standard curves prepared with the aid of pure specimens of these two substances, and the amount of vitamin A was calculated by subtracting from the total blue value, a blue value equivalent to the total carotene and xanthophyll found to be present. The results indicated that once the birds had been in production for some time the amounts of vitamin A, carotene and xanthophyll in the eggs depend chiefly on the diet. Yellow maize and green fodder, either fresh or dried, were the best sources of the vitamin A and carotene of the yolk. Yellow maize alone, constituting 25 per cent. of the ration, produced yolks with a vitamin A content of 150 I.U. per yolk, whilst the addition of lucerne resulted in even higher vitamin A contents, up to a maximum of 320 I.U. per yolk. A greater amount of xanthophyll than carotene was consistently found to be present, so that the yellow colour is chiefly due to the former. F. A. R.

Estimation of Riboflavin. I. New Biological Method. M. M. El Sadr, T. F. Macrae and C. E. Work. II. Estimation of Riboflavin in Milk: Comparison of Fluorimetric and Biological Tests. K. M. Henry, J. Houston and S. K. Kon. III. Statistical Analysis of the Data. J. O. Irwin. (*Biochem. J.*, 1940, 34, 601-612.)—Up to the present the biological assay of riboflavin has been hampered by the difficulty of providing adequate amounts of all the members of the vitamin B₂ complex other than riboflavin. The problem of providing such a supplement has now been solved by treating aqueous extracts of whole liver at pH 5 with Norit carbon; this removes all the riboflavin quantitatively, and the resulting filtrate contains adequate amounts of all other B₂ vitamins recognised to be essential for the rat. By means of this supplement a growth response curve to graded doses of riboflavin was constructed. By use of the curve assays have been made of the riboflavin-content of full cream

spray-dried milk and evaporated milk, and the results have been compared with those obtained by fluorimetric assays of the same samples of milk. One sample of spray-dried milk was found to contain 15.2γ of riboflavin per g. by the biological test and 10.0γ by fluorimetric assay. For another sample of spray-dried milk, the corresponding values were 9.3 and 10.3, and for an evaporated milk 2.6 and 2.9. The agreement was satisfactory when the milk was fed at a level not exceeding 10γ of riboflavin daily. F. A. R.

Estimation of Nicotinic Acid in Animal Tissues, Blood and Certain Foodstuffs. I. and II. E. Kodicek. (*Biochem. J.*, 1940, 34, 712-723, 724-735.)—The nicotinic acid contents of a wide range of animal tissues, foodstuffs and medicinal preparations were estimated by a colorimetric method almost identical with that described by Harris and Raymond (*cf. ANALYST*, 1940, 65, 183), involving the use of cyanogen bromide solution and *p*-aminoacetophenone. Some of the most important results obtained, expressed as γ per g. of fresh material, are as follows: liver (ox) 170, liver (sheep) 200, adrenals (sheep) 135, heart, muscle, kidney, pancreas, spleen, lung, brain (ox or sheep) 30 to 75, salmon 84, herring 40, cod 30, roe (herring or turbot) 21 to 23; liver extract (Eli Lilly "343"), 1090, marmite 640, yeast 74 to 91; egg-white <0.5, egg-yolk 10, milk (November to December) 0 to 5 (average 3), dried milk 25. F. A. R.

Combined Ascorbic Acid in Plant Foodstuffs. I. J. C. Pal and B. C. Guha. (*J. Indian Chem. Soc.*, 1939, 16, 481-495.)—Aqueous, alcoholic and ethereal extracts of cabbage, germinated kancha mung (*Phaseolus mungo*) and the Indian fruit bel (*Aegle marmelos*) were prepared. The ascorbic acid contents of the aqueous extracts were estimated by titration with 2:6-dichlorophenolindophenol before and after boiling in an atmosphere of nitrogen; it was found that the titration value was increased by the heat treatment. Even greater increases in the apparent ascorbic acid contents were observed when the alcoholic extracts were heated. With the ethereal extracts no reduction of the indicator occurred before heating, but an appreciable quantity occurred after heating. It was concluded that part of the ascorbic acid in these plant foodstuffs is present in a combined form ("ascorbigen"), from which the free vitamin is released by heating. By contrast, an alcoholic extract of mango showed a decrease in ascorbic acid content on heating, and therefore contained no "ascorbigen." The increased titration value observed after heating extracts of cabbage, mung and bel, cannot be attributed to the destruction of ascorbic acid oxidase as suggested by Van Eekelen; for the phenomenon was observed, first, with ethereal extracts that were shown not to contain the enzyme, and, secondly, with alcoholic extracts heated at 30 to 40° C. for 10 minutes, a temperature not sufficiently high to destroy the enzyme.

Ascorbigen can be obtained practically free from ascorbic acid by extracting the tissue with ether. It is readily hydrolysed by dilute acid at room temperature, and probably, therefore, by the gastric juice in the stomach.

II. P. N. Sen-Gupta and B. C. Guha. (*Ibid.*, 496-504.)—Dried cabbage was extracted with different solvents with the object of obtaining a concentrate of ascorbigen. Chloroform was found to be best for this purpose, and although by no means all the ascorbigen was extracted, the product was free from uncombined ascorbic acid. Dehydroascorbic acid was also absent, since treatment of the extract with hydrogen sulphide in the cold did not produce reducing substance. When the chloroform extract was evaporated and the residue extracted with water, all the ascorbigen was removed but only 20 per cent. of the total solids, so that this procedure effected a 5-fold concentration. This aqueous extract was used to test the effect of ascorbic acid oxidase on the reducing substances formed by heating ascorbigen. It was found that 60 to 70 per cent. disappeared on treatment with the enzyme, suggesting that, whilst the main product was ascorbic acid, some other non-specific reducing substance was also formed. Finally, the chloroform extract was fed to scorbutic guinea-pigs; the results showed not only that ascorbigen was active, but that its activity is comparable with that of an equivalent amount of ascorbic acid.

F. A. R.

Concentration of Ascorbigen from Cabbage. B. Ghosh and B. C. Guha. (*J. Indian Chem. Soc.*, 1939, 16, 505-510.)—The method previously used for obtaining a concentrate of ascorbigen (*cf.* preceding abstract) was not satisfactory, as the substance is not completely extracted by chloroform. Alternative methods were accordingly investigated, starting with cabbage juice. Glacial metaphosphoric acid produced a precipitate, but this contained only a small proportion of the ascorbigen together with some protein, whilst most of the ascorbigen remained in the filtrate. Other precipitants were tried with much the same result. The effect of various adsorbents was next tried, and it was found that charcoal adsorbed 60 per cent. of the ascorbigen from cabbage juice. The adsorbate was eluted by boiling for 1 hour with a 3 : 7 mixture of chloroform and absolute alcohol, and the pasty mass left after evaporation of the solvent contained combined ascorbic acid together with other material. The ascorbigen was further purified by extraction with a minimum quantity of water, centrifuging and evaporating the centrifugate to dryness in a desiccator. The product from 1 kg. of fresh cabbage contained the equivalent of 4.43 mg. of ascorbic acid. No further concentration could be effected by electro-dialysis, but some purification was effected by tungstic acid precipitation. The concentrate gave a strong Molisch reaction, reduced Fehling's solution, did not respond to the xanthoproteic, biuret or

Millon tests, but gave positive glyoxylic acid and Pauly reactions; it contained nitrogen and sulphur but no phosphorus. F. A. R.

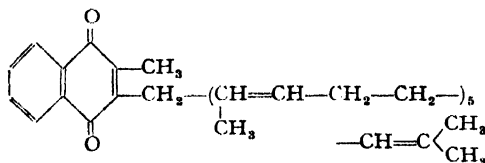
Estimation of Vitamin C in Foodstuffs. P. N. Sen-Gupta and B. C. Guha. (*J. Indian Chem. Soc.*, 1940, 16, 549-555.)—Trichloroacetic acid extraction followed by titration with 2 : 6-dichlorophenolindophenol tends to give a value for total vitamin C that is too low, because first some activity is lost by the action of ascorbic acid oxidase before this is destroyed by the trichloroacetic acid, and secondly, biologically potent ascorbigen does not reduce the dye unless split by heating. The use of mercuric acetate, as recommended by Emmerie and van Eekelen (*Biochem. J.*, 1934, 28, 1153), while eliminating interfering substances, causes some loss of ascorbic acid. The method now proposed consists in heating an aqueous suspension of the foodstuff in hydrogen sulphide followed by treatment with ascorbic acid oxidase. The enzyme was prepared according to the method of Tauber, Kleiner and Miskind (*J. Biol. Chem.*, 1935, 110, 211; *cf.* ANALYST, 1935, 60, 629), and purified by precipitation twice from aqueous solution by acetone; its potency was checked against pure ascorbic acid. Ten g. of the foodstuff were suspended in 50 ml. of water and treated with hydrogen sulphide on the water-bath. After removal of hydrogen sulphide with carbon dioxide or nitrogen, the mixture was treated with 2.5 ml. of 20 per cent. trichloroacetic acid. After centrifuging, the volume was made up to 100 ml. Ten-ml. aliquot portions were treated with 1 to 2 drops of sodium hydroxide solution to bring the pH to 5.6, followed by 2 ml. of *M* sodium acetate buffer solution (pH 5.6) and 3 ml. of the enzyme solution. The mixture was incubated at 40° C. for 30 minutes, made up to a definite volume and titrated against the dye solution. The difference in the titration value before and after incubation with the enzyme gave the amount of "true" ascorbic acid, comprising free ascorbic acid, dehydroascorbic acid and ascorbigen. This method was used for the estimation of ascorbic acid in 30 Indian foodstuffs, and in nearly every instance higher results were obtained by this method than by simple trichloroacetic acid extraction.

F. A. R.

Estimation of Vitamin D in Food Substances containing Phosphorus. K. H. Coward and E. W. Kassner. (*Biochem. J.*, 1940, 34, 538-541.)—It is well known that when rats are fed on a high calcium—low phosphorus diet, they develop rickets which can be largely, if not completely, healed by addition of phosphates to bring the Ca : P ratio more nearly equal to 1. Thus, unless the amount of phosphorus in the diet is relatively high, the effect produced by the addition of a foodstuff whose vitamin D-content is being estimated may be due partly to the vitamin D present and partly to the phosphorus. Experiments were therefore carried out to ascertain

the healing produced in rats fed on a rachitogenic diet of high calcium—low phosphorus content by (a) vitamin D alone, (b) phosphate alone, (c) vitamin D and phosphate together. The results indicate that the effect of giving both supplements is not additive, but multiplicative. It is concluded that the only practical method of assaying vitamin D in foodstuffs containing amounts of phosphorus sufficient to alter materially the Ca : P ratio is to saponify the material and carry out the assay on the unsaponifiable fraction. F. A. R.

Constitution of Vitamin K₂. S. B. Binkley, R. W. McKee, S. A. Thayer and E. A. Doisy. (*J. Biol. Chem.*, 1940, 133, 721–729.)—Vitamin K₂ is a yellow crystalline solid, m.p. 53.5 to 54.5° C., previously isolated from putrefied fish-meal. It has now been shown to be 2-methyl-1 : 4-naphthoquinone substituted in the 3-position by a long unsaturated chain which may be considered to be two farnesyl radicles fastened head to tail. Its structural formula is:



This constitution was established by the ozonolysis of the diacetate of dihydro-vitamin K₂, which yielded 1 : 4-diacetoxy-2-methylnaphthalene-3-acetaldehyde (identical with the compound obtained by the ozonolysis of the diacetate of dihydro-vitamin K₁), levulin-aldehyde and acetone. F. A. R.

Quantitative Test for Biotin and Observations regarding its Occurrence and Properties. E. E. Snell, R. E. Eakin and R. J. Williams. (*J. Amer. Chem. Soc.*, 1940, 62, 175–178.)—An assay method is described for biotin; it depends on the growth of a strain of *Saccharomyces cerevisiae*. The yeast was grown on a basal medium consisting of sucrose 20 g., ammonium sulphate 3 g., potassium hydrogen phosphate 2 g., magnesium sulphate (hydrate) 0.25 g., calcium chloride (hydrate) 0.25 g., boric acid 1 mg., zinc sulphate 1 mg., manganese chloride 1 mg., titanous chloride 1 mg., ferric chloride 0.5 mg., copper sulphate (hydrate) 0.1 mg., potassium iodide 0.1 mg., L-aspartic acid 0.1 g., inositol 5 mg., β-alanine 0.5 mg., aneurin 20γ, and vitamin B₁₂ 20γ, dissolved in 1 litre of distilled water. Two ml. of an aqueous solution of the material to be tested were sterilised and cooled, and 10 ml. of the basal medium were added containing 0.02 mg. of suspended yeast. The mixture was incubated at 30° C. for 16 hours and then diluted with 10 ml. of saturated aqueous chlorothymol solution and shaken for 3 to 5 minutes. The amount of yeast present was estimated by means of a thermocouple and galvanometer (Williams, McAlister and Roehm,

J. Biol. Chem., 1929, 83, 315; Abst., ANALYST, 1929, 54, 613), this method being found superior to measurement by means of a photoelectric colorimeter. Blank cultures and cultures containing known amounts of biotin (0.000025 to 0.00025γ) were run simultaneously with the unknowns, and the amounts of biotin in the unknown samples were read off from a standard curve. It was found that in parallel tests of a crude liver extract and a biotin solution, curves of identical character were obtained when the concentration was plotted against the weight of yeast produced; it is concluded that the test is highly specific for biotin. Vitamin B₁₂ must be present in the medium before any significant effect of biotin is observed. The following results (in γ per g.) were obtained, clear aqueous extracts being produced by autoclaving the material for 10 minutes at 15 lb. pressure with a large volume of water, except that casein was hydrolysed with sulphuric acid: cane molasses 1.7 to 2.1, beet molasses 0.06 to 0.9, whey 0.12, egg white 0.05, egg yolk 0.37, yeast 0.13, casein 0.125, potato 0.01, autolysed liver 3.9. F. A. R.

Bacteriological

Thermal Death Time of Micro-organisms encountered in the Brewing Industry. S. S. Epstein and F. D. Snell. (*J. Inst. Brewing*, 1940, 175–178.)—Twenty-one cultures were studied, including 5 *sarcina* (*Pediococcus*) strains, 2 *Lactobacillus* strains, 2 *Acetobacter* strains, 8 yeasts and 4 cultures from infected bottled beer, storage ale, storage beer and draught beer. The pure strains of bacteria were cultivated in sterile beer at 77° F. until turbidity developed and then kept at room temperature for six days before testing. The yeasts were grown in brewers' wort under the same conditions. The object of using cultures 6 days old was to simulate practical conditions, the cultures having by this time attained maximum resistance to heat. The size of the inoculum was 10,000 to 30,000 organisms per ml. The results of these studies indicate that the standard practice of pasteurisation at 140° F. for 20 minutes should render beer free from all living organisms commonly encountered in the brewing industry. The order of heat resistance from the greatest to the least was found to be:—*Lactobacillus*, *Pediococcus* (*Sarcina*) and *Acetobacter*. None of the bacteria tested was able to survive an exposure to 60° C. (140° F.) for ten minutes. Of the yeasts, the order of heat resistance was:—*S. pastorianus*, *S. ellipsoideus* and brewers' culture yeasts, *Mycoderma* and *Torula*. Yeast spores were killed at 58° C. (136.4° F.) within 20 minutes and at 60° C. within 15 minutes. D. R. W.

Mechanism of the Microbial Oxidation of Ammonia. Part II. G. Gopala Rao and W. V. Sundara Rao. (*J. Indian Chem. Soc.*, 1939, 16, 681–690.)—The inhibitory action of capillary active substances such as

the urethanes, nitriles, alcohols and ketones, and of the cyanides on the microbiological oxidation of ammonia has been investigated. The tests were made in flasks containing 100 ml. of culture medium, one ml. of enriched culture of the nitrite-forming bacteria, and varying volumes of the test substance; the whole was made up to 200 ml. and portions of the liquid were withdrawn from time to time for the determination of nitrites. The tabulated results show that these substances inhibit the respiration of the bacteria for a time, but that ultimately nitrite begins to appear, the length of time before its appearance increasing with the concentration of the added narcotic. These experiments lead to the conclusion that oxidation of ammonia is a surface catalytic reaction taking place at certain active centres on the surface of the bacteria. Recent work of Quastel and Woolbridge shows that a suspension of bacteria behaves in a manner identical with any colloidal system possessing catalytic properties, and the fact that resting bacteria are able to bring about reversible reactions whose equilibrium points are independent of the amount or conditions of the organism supports this view. Some bacteria, such as *B. coli*, have a wide range of activation, effective for as many as 56 substances. According to Quastel this is associated with specific patches on the surface of the bacteria, which first adsorb and then activate the substance. The more elementary nitrifying bacteria cannot utilise any of the complex organic molecules; they oxidise ammonia and make use of the exothermic reaction for their life activities and for the chemo-synthesis of carbohydrates from atmospheric carbon dioxide. Whereas *B. coli*, *pneumococci* and *staphylococci* are presumed to have a variety of specific patches on their surface the nitrifying bacteria are presumed to have only one kind. In the author's opinion the inhibitory action of urethane, alcohols, etc., is not specific and these so-called poisons act by competing with the substrate for the space available for adsorption. With the cyanides, however, the resulting inactivation is presumed to be due to the centres containing an iron-rich complex, the function of which is completely inhibited by combination with the CN' ion. The results here recorded show that in the same series of compounds the concentration required for marked inhibition is lower with compounds of higher molecular weight. Thus with ethyl alcohol it is $M/10$ with acetone $M/20$, with octyl alcohol $M/500$ and with benzophenone $M/4000$.

D. R. W.

Gas Analysis

Starch-iodide Method for Ozone. C. E. Thorp. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 209.)—Many of the reagents proposed as substitutes for potassium iodide (*cf.* Benoist, *ANALYST*, 1919, 44, 183; Briner and Perrottet, *Helv. Chem. Acta*, 1937, 20, 293;

Abst., ANALYST, 1937, 62, 415) have been found satisfactory, but potassium iodide is preferred as a quick and convenient method for the detection of ozone. When ozone in admixture with air was passed through neutral potassium iodide solution and the iodine liberated on acidification was titrated with standard sodium thiosulphate solution the greatest sensitivity obtained was the detection of 0.0013 mg. of ozone per ml. of 2 N potassium iodide solution. Since analyses of air containing 0.1 p.p.m. by weight of ozone are frequently required, 9.9 litres of air would have to be passed through each ml. of potassium iodide solution before the ozone could be detected in this way. Various methods have been tried to increase the sensitivity of the potassium iodide reagent (Baskerville and Crozier, *J. Amer. Chem. Soc.*, 1912, 34, 1332; *Abst., ANALYST*, 1912, 37, 587; Ernst, *Biochem. Z.*, 1931, 232, 346). The use of thiocyanate ions (Ernst, *loc. cit.*) may increase the sensitivity towards some compounds, but appears to decrease it towards ozone. The use of free acid to lower the pH of the potassium iodide solution introduces another error, owing to the formation of hydrogen peroxide. The use of a buffer solution greatly increased the sensitivity without introducing the error caused by free acid. Five ml. of an aqueous solution of 5 g. of aluminium chloride hexahydrate and 1 g. of ammonium chloride per litre are added to each 100 ml. of potassium iodide solution, and the mixture should not be acidified during the subsequent titration. The sensitivity of the mixture is 0.00062 mg. of ozone per ml. of potassium iodide solution and the sensitised solution is stable for over three hours. The ozone sample is drawn through 100-ml. gas-washing bottles each fitted with a Jena glass disc (Thomas, *Ind. Eng. Chem., Anal. Ed.*, 1933, 5, 193) until a deep colour of iodine is formed in the first bottle. The sodium thiosulphate solution for the titration should not exceed 0.01 N, and a 2 ml. micro-burette is recommended for greater accuracy. For ozone concentrations of less than 0.5 p.p.m. the gas-washing bottles should be of the semi-micro type, and the volume of the test solution should not exceed 10 ml. There must be no exposed cork or rubber stoppers; ground glass connections are preferable, but neoprene or rubber or cork coated heavily with shellac or lacquer may be used. Only ultra-violet light will produce pure ozone. The product of other types of commercial ozoniser will contain hydrogen peroxide and oxides of nitrogen. To remove these the ozone should be passed through absorption tubes containing chromic acid and potassium permanganate.

A. O. J.

Determination of Carbon Disulphide in Air. F. F. Moorhead. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 373-374.)—A copper-diethylamine reagent is used, *viz.* 1 ml. of a 1 per cent. solution of diethylamine in 2-methoxyethanol mixed with 2 drops of 0.1 per cent. cupric acetate solution in 2-methoxy-

ethanol. A yellow or brownish colour is produced with carbon disulphide. The reaction is more sensitive and more suitable for colorimetric work than the xanthate reaction. Methoxyethanol is better than ethanol as reaction medium, as the colour is more stable. The method involves drawing a sample of air (100 ml. is usually sufficient) through the reagent. A suitable absorption tube consists of a 6 to 7 cm. length of glass tubing, 10 mm. in diameter, containing glass beads which are retained by constrictions at the ends. The tube is used in a vertical position, the lower end being sealed to an upturned length of capillary tubing in which a bulb is blown, and the upper end sealed to a right-angled leading tube. The reagent is placed in the absorption tube connected at the upper end to an aspirator, which conveniently consists of a tap funnel containing water. After aspiration, the contents of the absorption tube are rinsed into a 2-ml. measuring flask and made up to a definite volume with the solvent. For the colorimetric comparison, which is made after about 20 minutes in a Klett (Duboscq type) colorimeter with 2-ml. cups, a standard solution of carbon disulphide in methoxyethanol is added to the reagent. The method is capable of determining 1 to 30% of carbon disulphide. If hydrogen sulphide is present the air should be passed through 5 per cent. aqueous copper sulphate or lead acetate paper and then through a desiccating agent before entering the carbon disulphide absorption tube. S. G. C.

Agricultural

Electrodialysis of Soils. Influence of Exchangeable Bases on the Recovery of Manganese by Electrodialysis. R. C. Hoon and C. L. Dhawan. (*J. Indian Chem. Soc.*, 1940, 17, 195-204.)—Recovery of manganese from soils by electrodialysis is affected by the presence of other bases in the exchange complex. Three natural soils were freed from exchangeable bases and converted into manganese soils (*cf.* Prince and Toth, *Soil Science*, 1938, 46, 83). The manganese-contents of the artificial soils were determined (i) directly by the bismuthate method, (ii) in the leachates, obtained from the soils with *N*-neutral ammonium acetate solution, and (iii) by electrodialysis for 5 hours. Sets of (manganese + base) soils were prepared from each artificial soil by addition of different amounts of sodium, potassium, calcium and magnesium hydroxides. The soils were then shaken with water for 48 hours and electrodialysed. Results were tabulated for base added to soil, pH of 1 : 5 suspensions, and percentage recovery of the other base. A few natural soils, from good and bad areas, containing different amounts of manganese were also electrodialysed for 5 hours. Manganese was determined separately in the electrodialysate recovered in the first hour and in the collective electrodialysate of the next four hours; the total recovery in 5 hours and the percentage of total manganese in it were calculated. In the artificial

soils, the manganese-sodium sets yielded the maximum amount of electrodialysable manganese and the minimum amount as deposit on the cathode, etc. In these and in the manganese-potassium sets there was on the whole a tendency for the manganese recovered in the deposit to decrease with increasing pH. In the manganese-magnesium sets the least manganese was recovered in the electrodialysate and mainly in the deposit. In the manganese-calcium deposit on the cathode, the manganese-content decreased as the pH increased to about 8.0, then increased slightly. The results showed that soils with high pH values and high manganese contents yield more manganese on electrodialysis than those with low pH values and low manganese contents; all, or almost all, the electrodialysable manganese is obtained in the first hour. It is concluded that at high pH values when the dominant base is sodium, more manganese comes out in the soluble form and is thus available to plants; this availability may be reduced when the dominant base in the exchange complex is calcium or magnesium.

E. B. D.

Determination of Rotenone in Derris Root. T. M. Meijer and D. R. Koolhaus. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 205-209.)—Analyses of the same sample of powdered derris root by different workers showed that differences of about 4 per cent. in the nominal value of the rotenone-content of the dry sample and large differences in the moisture-content may occur. A study of the methods proposed for the determination of rotenone in derris root showed that several gave results concordant with those obtained by the method here described. Methods in which the extraction is complete and sufficient attention is paid to the crystallisation of the rotenone or rotenone and carbon tetrachloride solvate, should give similar results. Suitable solvents for extraction are ether, benzene, chloroform, ethyl acetate and trichloroethylene. Carbon tetrachloride should not be used, especially with samples of high rotenone-content, as Seaber (*J. Soc. Chem. Ind.*, 1937, 56, 168T; *Abst., ANALYST*, 1937, 62, 630) and Jones and Sullivan (*J. Econ. Entomol.*, 1938, 21, 148) have shown. At least 75 per cent. of the powdered root must pass through an 80-mesh sieve. The powder (50 g.) held in a Soxhlet extractor without a thimble by means of a cotton-wool plug, was extracted with ether for at least 65 hours. The ether was distilled off in a 100-ml. centrifuge tube, and any rotenone separated during the extraction was transferred to the tube and the flask was rinsed out with ether until the liquid in the tube measured 25 ml. The tube was kept at room temperature for a day and in a refrigerator for a further 2 days. The mother liquor was poured into a flask, the remaining rotenone was broken up with the addition of 10 to 15 ml. of ether, and the tube and flask were placed in the refrigerator for another day. The tube was centrifuged at 3500 r.p.m. for

3 to 5 minutes and the supernatant liquid was added to the mother liquor. The centrifuge tube was then dried at 70° C., and finally at 100° C. *in vacuo*, and its contents were weighed. The purity of the rotenone was ascertained by means of an empirical table correlating its melting-point with its purity. If the m.p. was below 140° C. the product was centrifuged with 10 ml. of ether and its m.p. again determined. Sometimes substances with m.p. very near that of rotenone separate (Meijer and Koolhaus, *Rec. Trav. Chim. Pays-Bas*, 1939, 58, 207), and for this reason the purity of the product was ascertained also from its optical rotation (Seaber, *loc. cit.*). For each ml. of the mother liquor and washings 4.2 mg. of rotenone were added as a correction for solubility, and any additional rotenone that separated from the mother liquor was added to the crude rotenone. The mother liquor was evaporated in a 500-ml. tared round-bottomed flask on the water-bath and finally *in vacuo* at 40° C. The residue was dried over lime for 2 days and weighed, and the weight of resin so obtained when added to the weight of crude rotenone gave the total ethereal extract. Moisture was determined by heating 2 to 3 g. of the powdered sample to constant weight at 105° C. To determine unextracted rotenone the resin was treated first with boiling petroleum spirit and then with boiling cyclohexane, and the rotenone was separated from the residue as the carbon tetrachloride solvate. An amount corresponding with about 9 per cent. of the rotenone originally found was recovered from the resin, especially when the sample had a high ethereal extract and a low rotenone-content. Determination by a chromatographic method gave a similar result. Jones and Graham (*J. Assoc. Off. Agr. Chem.*, 1938, 21, 148) have proposed a method in which the ground root is extracted with chloroform and the rotenone determined by conversion into the carbon tetrachloride solvate. Extraction with chloroform at room temperature is practically complete. The value for pure rotenone found by the ether-extraction method was almost equal to that found for crude rotenone by the method of Jones and Graham, and, as the solvate obtained in the latter method is fairly pure, the value should not be lowered by a correction for purity. A satisfactory uniform method might be based on the Jones-Graham method, but it must be adapted to samples in which the ratio of rotenone to total extractive is high. This may be effected by grinding samples so that at least 75 per cent. passes through a 200-mesh sieve, or by extracting the powdered root several times with chloroform. Jones (*Ind. Eng. Chem., Anal. Ed.*, 1937, 9, 206) has determined rotenone by conversion of the carbon tetrachloride solvate into the dichloroacetic acid solvate. Since each molecule of dichloroacetic acid is associated with a molecule of rotenone, the solvate may be titrated with standard alkali solution. Derris root ground to pass a 200-mesh sieve lost a considerable amount of its apparent rotenone-content when heated to 60° C. and

still more at 80° C. Heating the sample to reduce the moisture-content before analysis is therefore to be discouraged. A. O. J.

Organic

Rapid Qualitative Test for Alcoholic Hydroxyl Groups. F. R. Duke and G. F. Smith. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 201-203.)—The red colour of solutions of hexanitrate or hexaperchlorate potassium or ammonium cerate in alcohol suggested the use of these substances as reagents for the detection of the alcoholic hydroxyl group. A solution of 400 g. of hexanitrate ammonium cerate in a litre of 2 *M* nitric acid is approximately 0.7 *M* in nitrate cerate ion. A solution of perchlorato ceric acid in 6 *M* perchloric acid is available commercially and is approximately 0.5 *M* in perchlorato cerate ion; it is prepared by electrolytic oxidation of cerous perchlorate in perchloric acid solution. With this solution (unlike the hexanitrate solution) the concentration of the acid is not an important factor. The procedure is as follows:—The cerate reagent (1 ml.) is diluted with 2 ml. of water, and 1 or 2 drops of the compound to be tested (or its saturated aqueous solution) are added. A red colour indicates an alcohol. In testing compounds insoluble in water 1 ml. of the cerate reagent is mixed with 2 ml. of dioxane, and 1 or 2 drops of the compound (dissolved if necessary in the least amount of dioxane) are added. Dioxane solutions cannot be used with the perchlorato cerate reagent owing to reduction of the reagent. Acids, aldehydes, ketones, esters and hydrocarbons do not interfere with the test. Amines raise the pH to such an extent that insoluble cerium compounds may be precipitated. Aromatic amines and phenols interfere by forming characteristic colours and precipitates. Easily oxidised substances (*e.g.* oxalic acid) interfere by reduction of the reagent, and organic dyes may cause colour interference. Alcohols containing up to 10 carbon atoms are included in the test. Primary, secondary and tertiary alcohols give instantaneous reactions. Hydroxycarboxylic acids also react. With glucose, sucrose and dextrin the colour is rapidly lost by oxidation. Solutions containing 2 to 4 per cent. of butyl alcohol react with the nitrate cerate reagent, and solutions of 1 to 2 per cent. with the perchlorato cerate reagent. The more reactive the alcoholic hydroxyl group the more sensitive the test; *e.g.* benzyl and allyl types are especially sensitive. The oxidation potential of the nitrate cerate reagent is much lower than that of the perchlorato cerate reagent (Smith and Getz, *Ind. Eng. Chem., Anal. Ed.*, 1938, 10, 191). For this reason the colour with the nitrate reagent is more permanent. Tertiary alcohols form much more stable coloured solutions than primary and secondary alcohols, and this may serve as a means of differentiation. The colour obtained with the four butyl alcohols was studied quantitatively in a photoelectric

colorimeter. The intensity was found to increase progressively with the four isomeric alcohols in the order iso-, secondary, normal, tertiary. In all instances the colour was more intense with the perchlorate cerate reagent and more stable with the nitrate cerate reagent, and was most pronounced with the tertiary alcohol. By comparison of methyl alcohol with nonyl alcohol the intensity of colour was shown to diminish with the longer chain. The colour produced by aqueous solutions of methyl alcohol is greater than that produced by solutions in dioxane. The use of perchloric acid with organic compounds in the manner described introduces no danger in the application of the test provided that the solutions are not heated. A theoretical explanation of the colour formation is given.

A. O. J.

New Reaction of Formic Acid. T. L. Davis and W. P. Green. (*J. Amer. Chem. Soc.*, 1940, 62, 1274-1276.)—Interaction of bromine and formic acid produces a substance which, in presence of bromine, reacts with aniline to form derivatives of urea. A mixture of 12 g. of bromine and 30.7 g. of anhydrous formic acid was kept for 2 days, then run slowly from a separator into a solution of 108.3 g. of aniline in 300 ml. of benzene, which was mechanically stirred. The solution (which contained a precipitate) was made alkaline, steam distilled to remove aniline and brominated aniline, and extracted with ether. On evaporation, the extract left 0.2644 g. of a crude product which on fractional sublimation yielded impure carbanilide and a brominated carbanilide fraction. Treatment of the second fraction with aniline followed by sublimation and repeated recrystallisation also yielded carbanilide. It is considered probable that dibromodihydroxy-methane is formed in presence of excess of bromine, by addition of bromine to the tautomeric form of the acid—dihydroxy bivalent carbon.

E. B. D.

Separation of Hydroxy from Non-hydroxy Fatty Acids by means of a Dibasic Acid Anhydride. F. E. Kurtz and P. S. Schaffer. (*J. Amer. Chem. Soc.*, 1940, 62, 1304-1305.)—Separation of hydroxy fatty acids from non-hydroxy acids by the usual methods is difficult when the differences in their physical properties, or in those of their derivatives, is small. A better method is to heat the mixed esters of the mixed acids with a dibasic acid anhydride, dissolve the reaction mixture in petroleum spirit and extract the derivative of the hydroxy ester with alkali. Owing to its solubility in esters, maleic anhydride is recommended for separating hydroxy acids from saturated acids, but with unsaturated acids partial saturation occurs. For these, succinic acid may be used, but it requires addition of a solvent; with dioxane low yields are obtained; pyridine gives better results but forms a tarry precipitate on prolonged heating, though much less than with maleic anhydride. With either reagent more than one separation is usually required. With castor oil acids

(about 85 per cent. of hydroxy acids) two separations give hydroxy acids practically free from non-hydroxy acids.

E. B. D.

Purification of High Molecular Weight Fatty Esters. L. O. Buxton and R. Kapp. (*J. Amer. Chem. Soc.*, 1940, 62, 986.)—The following procedure is recommended for removing substantially all of the fatty acids that have not reacted in the preparation of their esters. The alcohol is distilled from the esterification mass, and the residual mixture of free fatty acids, crude ester and catalyst is dissolved in 2 to 5 parts by weight of solvent such as ethylene dichloride to 1 part of ester. Ten to 20 g. of the solution are dissolved in a mixture of alcohol and ether and titrated with standard 0.5 N alcoholic potassium hydroxide solution. On the basis of this titration value an equivalent weight of concentrated (preferably 38 per cent.) aqueous potassium hydroxide solution is added slowly, with constant stirring, to the ester solution. The potassium salts of the fatty acids that have not reacted and of any mineral acid present as catalyst are filtered off without suction and washed with a small quantity of fresh ethylene dichloride to remove traces of neutral ester. The filtrate is distilled without drying. The esters so prepared have acid values of 0.5 to 1.0 and can be further purified by vacuum distillation. The method has been used to obtain quantitatively the yields of methyl and ethyl esters of lauric, oleic, linolic, stearic and ricinoleic acids from the original esterified mass prepared with the respective crude acids, and also for the preparation of relatively pure mono- and dinaphthenates of diethylene glycol.

Thiocyanogen Value of Linolenic Acid. J. P. Kass, H. G. Loeb, F. A. Norris and G. O. Burr. (*Oil and Soap*, 1940, 17, 118-119.)—The thiocyanogen value of α -linolenic acid was studied under the conditions of the determination recommended by the American Oil Chemists' Society (*Ind. Eng. Chem., Anal. Ed.*, 1936, 8, 233). The thiocyanogen solutions, which varied in concentration from 0.16 to 0.18 N, were freshly prepared for each series of determinations and were dispensed from a 25-ml. automatic pipette protected with a calcium chloride tube. All glassware was dried for one hour at 105° C., and the reaction temperature was maintained at 19° \pm 1° C. by keeping the reagents and reaction mixtures in a large water-jacketed incubator. Ethyl linolenate (iodine value, Wijs, 247.3; theory 248.5) was prepared by debromination of purified hexabromostearic acid (from linseed oil) with zinc and 7.5 N ethyl alcoholic hydrogen chloride according to the method of Rollet (*Z. physiol. Chem.*, 1909, 62, 422). In determinations in which the excess of 0.16 N thiocyanogen solution used varied between 150 and 290 per cent., whilst the time of contact was either 20, 24 or 28 hours, it was found that the thiocyanogen value of ethyl linolenate approximated to 151.2, corresponding with 167.3 for linolenic acid (of theoretical

iodine value 273.7) instead of 182.5 as assumed by Kaufmann and Keller (*Z. angew. Chem.*, 1929, 42, 73). Experiments on mixtures of corn (maize) oil acids with varying proportions of α -linolenic acid (28–30 hours' contact with the 0.16 *N* reagent) showed that the observed thiocyanogen values agreed with those calculated on the assumption that the linolenic acid has a thiocyanogen value of 167.3 and not 182.5, subject only to slight deviations from the calculated values with increasing concentrations of linolenic acid in the mixed acids. The authors suggest the substitution of the empirically determined value of 167.3 for the thiocyanogen value of linolenic acid in equations used in calculating the proportion of this acid by thiocyanometric analysis. The empirical value is also shown to vary somewhat with the concentration and excess of the thiocyanogen reagent and with the period of its contact with the unsaturated acids or esters (*cf.* Hilditch and Murti, *ANALYST*, 1940, 444, footnote). T. P. H.

Separation and Characterisation of Petroleum Acids. H. G. Schultze, B. Shive and H. L. Lochte. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 262–266.)—Preliminary investigation of alkali wash from the refining of light burner oil showed that of the numerous combinations of physical constants suggested for the rapid characterisation of fractions the product of the refractive index and the density was the simplest and most useful. Calculation of the value of $n_D^{20} \times d_4^{20}$ for a large number of reported aliphatic, unsaturated and naphthenic acids, phenols and hydrocarbons showed that the aliphatic acids have products ranging from 1.280 to 1.350 with most values between 1.300 and 1.310, the naphthenic acids products range from 1.390 to 1.470 with most values between 1.410 and 1.440, the products of phenols have values above 1.500, and those of hydrocarbons values below 1.300 and usually below 1.280. The only exception found among the acids containing the cyclopentyl group—the typical naphthenic acids—was 1,2,3-trimethylcyclopentane carboxylic acid (Noyes and Burke, *J. Amer. Chem. Soc.*, 1912, 34, 174). Its constants yield the value 1.313, which is within the range of the typical aliphatic acids. The acids liberated from a large quantity of conc. alkali were distilled under reduced pressure and the fractions so obtained were submitted to further fractionation. The fraction of the desired boiling range was separated into strong and weak acid components by fractional neutralisation, and further separation was effected by fractional neutralisation of the strong acids with sodium carbonate. Repetition of this procedure with various non-phenolic fractions effected the separation of the strongest organic acids, which were selected for more detailed study. During some of the later fractionations a solid substance separated in the condenser, and this was ultimately identified as dimethyl maleic anhydride which has since been isolated from a separate source (Hancock and Lochte, *J. Amer. Chem. Soc.*,

* 1939, 61, 2448). The following procedure for the separation of acids by means of their silver salts was found satisfactory:—The silver salts were fractionally precipitated in the usual manner, each fraction was dissolved in ammonia, and the solution was fractionally neutralised with *N* nitric acid. When the acids from the first fractions were liberated from their silver salts by treatment with dehydrated phosphoric acid and subsequent distillation, a main fraction of dry colourless acids was obtained. By repetition of the silver salt separations a pure compound was obtained, and, by analysis of its *o*-phenylenediamine and *p*-nitrobenzyl derivatives, it was ultimately identified as *n*-valeric acid. By similar methods *n*-butyric acid, caprylic acid and *n*-octanoic acid were isolated and identified. When the final filtrate from the silver salt method was acidified and steam-distilled the presence of *o*-nitrophenol was detected. Although the yield was small, the formation of this substance was interesting because the absence of phenol in petroleum acids has often been reported. Fractions for which the product of density and refractive index indicated the presence of naphthenic acids were selected and submitted to further silver salt separations and conversion into acyl chloride and amido derivatives. Finally the amides of *p*-hexahydrotoluic acid and some of its isomers were isolated and identified. Although formerly the naphthenic acids in petroleum were thought to consist of cyclohexyl acids, the tendency in the last decade has been to assume that naphthenic acids are all cyclopentyl acids, since only very little indication of cyclohexyl acids had been obtained and none had been isolated. The amount of the pure solid isomer of *p*-hexahydrotoluic acid obtained in this investigation was very small, but other isomers appeared to be present in larger amounts. Since the acids isolated were obtained from a mixture of straight run and cracking process products, the cyclohexyl acids may have been formed during the cracking process. A. O. J.

Colorimetric Determination of Primary Mononitroparaffins. E. W. Scott and J. F. Treon. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 189–190.)—When samples of air containing nitroethane were passed through dilute sodium hydroxide solution and the alkaline solution was reduced by means of iron, tin or zinc after acidification with sulphuric acid, quantitative yields of ethylamine were not obtained, although the reduction of aqueous solutions of nitroethane was satisfactory. When the alkaline solutions were added to an excess of hydrochloric acid containing ferric chloride a pink colour was formed, and a method of determining as little as 0.5 mg. of nitroethane in 25 ml. was based on this reaction. An aliquot portion of the solution, containing 1 to 20 mg. of nitroethane in 1 to 15 ml., is neutralised and treated with 1.5 ml. of 20 per cent. sodium hydroxide solution in a 25-ml. flask. After 15 minutes the solution is acidified with 6 ml. of dilute hydrochloric acid

(1 + 7) and 0.5 ml. of 10 per cent. ferric chloride solution is added immediately. Comparison solutions containing approximately the same amounts of nitroethane are treated similarly. After standing for 15 minutes the solutions are made up to 25 ml. and the colours are compared. A colorimeter equipped with a 1.58 cm. Wratten filter No. 65A in B glass (Eastman Kodak Co.) is recommended for making the comparison. The optimum pH for the greatest intensity and stability of the colour is 1.25 to 1.30. Solutions containing more acid fade gradually, and those with a higher pH do not change from the initial brownish colour to the final deep red. With very small amounts of nitroethane colour interference is prevented by reducing the amount of ferric chloride solution. The colour should always be developed at room temperature; it fades rapidly when the solution is heated. Nitroethane solutions which have not been made alkaline do not react with the ferric chloride, and alkaline solutions allowed to stand for a short time after acidification no longer react. Apparently only the aci-form of the nitro-compound gives the reaction. Stable coloured complexes with ferric chloride are also formed by 1-nitropropane and 1-nitrobutane, and colorimetric methods have been based on these reactions. Ferric chloride reacts with 2-nitropropane and with 2-nitrobutane, but the colours fade rapidly. Nitromethane does not react but may be determined by means of its colour reaction with vanillin and ammonia (Manzoff, *Z. Unters. Nahr. Genussm.*, 1914, **27**, 469; *Abst.*, *ANALYST*, 1914, **39**, 264). The complex formed with nitroethane was isolated but proved too unstable for purification and analysis. Spectrophotometric examination of the coloured solutions showed that for the ranges of concentration used in the test the absorption followed the Lambert-Beer law. The method gave satisfactorily accurate results with samples of air containing known amounts of nitroethane, and was also successfully applied with slight modifications to the determination of nitroethane in tissues. A. O. J.

Identification of Organic Compounds.

II. Piperidyl Derivatives of Aromatic Halogenonitro Compounds. M. K. Selkel.

(*J. Amer. Chem. Soc.*, 1940, **62**, 750-756).—The characteristics of the piperidyl derivatives of aromatic halogenonitro compounds and the rate of formation of the derivatives can be used to identify the aromatic bodies. The two following general procedures were worked out: (A) To 0.5 g. of the halogenonitro compound add 1.5 ml. (1.0 ml. for bromo compounds) of piperidine. Observe phenomena such as spattering (stir to prevent local over-heating), boiling, colour formation, and the time elapsing before piperidine hydrohalide separates; these are partly dependent on the quantities used. Heat the resulting solution or mixture under reflux in an oil-bath for one hour, cool, add water and filter. If the product separates as an oil, the following crystallisation methods

should be tried in order: (a) place the mixture in ice for an hour with scratching (with few exceptions this method is sufficient); (b) decant the aqueous layer containing excess piperidine and piperidine hydrohalide and wash the oil several times with water; (c) freeze in dry-ice. If no crystallisation is obtained, abandon the experiment. The yields are 90 to 100 per cent. and the crude product will melt sharply if the chlorine replacement is complete and if decomposition products due to excessive reactivity are not present. Recrystallise from 85 per cent. alcohol, using excess if the m.p. is low. Certain high-melting insoluble compounds, which are indicated in tables, require 95 per cent. alcohol. The yield of piperidine hydrohalide, which is an index of the completeness of the reaction, can be determined qualitatively by evaporating the aqueous filtrate of the crude product and weighing the residue. If a nitro group is replaced, the residue is a reddish acetone-soluble gum, giving a nitrosamine reaction with diphenylamine reagent. (B) Dissolve 0.5 g. of the compound in 5 ml. of boiling alcohol (more if necessary), add 1.5 ml. (1.0 ml. for bromo-compounds of piperidine) and heat for fifteen minutes on the steam-bath. If the product does not separate on cooling or icing, force it out with water, crystallising any oil obtained as described under procedure A. If water has not been added before filtering, wash the precipitate with water to remove possible contaminating piperidine hydrohalide. The yields are only 70 to 90 per cent., but the crystallisation from alcohol effects purification. The times of reaction in both of these procedures may be altered to suit individual materials, and the following procedures may also be applied. (C) Mix the two reactants and add water after a definite time (which is indicated for those substances studied). (D) Add ice-cold piperidine to the solid compound (also iced), continue icing until the initial violent reaction has moderated and then allow the mixture to stand in a water-bath at room temperature, stirring frequently, until the spontaneous evolution of heat ceases (1 to 2 hours). (E) Dissolve the compound in the minimum quantity of cold alcohol (20 to 40 ml. for 0.5 g.), add piperidine and after the specified time at room temperature add water to precipitate the product. To prepare monopiperidyl derivatives when dipiperidination is rapid exactly two mols. of piperidine must be used. Thirty-seven halogenated nitrobenzenes were studied, and the conditions for the preparation of their piperidyl derivatives are given. Proofs of the identity of certain of the derivatives are given.

E. M. P.

New Colour Reaction for Diarylamines.

E. M. Meade. (*J. Chem. Soc.*, 1940, 1808).—The amine is dissolved in a little anisole and a solution of methylmagnesium iodide in anisole is added, followed by benzoyl chloride. If a diarylamine were originally present a pronounced red colour is produced. The test is particularly useful for following the course of

N-substitution of diarylamines; for example, it will detect 1 per cent. of unchanged diphenylamine in a sample of N-methyldiphenylamine. The following substances gave a positive reaction: diphenylamine, 4'-methoxy-4-methyldiphenylamine, 4:4'-dimethoxydiphenylamine, phenyl- β -naphthylamine, *o*- and *p*-methoxyphenyl- β -naphthylamines; the following failed to give the reaction: N-methyldiphenylamine, N-methyl-, -acetyl-, and -benzoyl-4:4'-dimethoxydiphenylamines, aniline, methylaniline, dimethylaniline, benzyaniline, and *p*-anisidine.

E. M. P.

Estimation of Lignin in Tannin Materials. J. G. Shrikhande. (*Biochem. J.*, 1940, **34**, 783-789.)—When the lignin present in plant tissues was estimated by the 72 per cent. sulphuric acid method after pre-treatment with 5 per cent. sulphuric acid, surprisingly high results were obtained with certain types of materials, such as tea leaf prunings. It appeared as though the pre-treatment, instead of removing extraneous material that interfered with the lignin estimation, rendered it insoluble. The high results obtained coincided with the presence in the materials of tannins, especially of catechol tannins, which are precipitated by acids and thus rendered insoluble. A modified method of pre-treatment, to free the material from tannins, caffeine, chlorophyll, proteins and pentoses, all of which were shown to interfere with the estimation, was worked out. Two g. of the oven-dry material (powdered to pass an 80-mesh sieve) are extracted in a Soxhlet apparatus with 200 ml. of 95 per cent. alcohol for 4 hours. The residue is boiled under reflux with 150 ml. of water for 1 hour, the extract is filtered off, and the residue is hydrolysed with 150 ml. of 5 per cent. sulphuric acid for 1 hour. The product is filtered off on a weighed sintered glass crucible, washed free from acid, dried and weighed. Portions (0.2 g.) of this material are weighed into 1-litre beakers and treated with 20 ml. of 72 per cent. sulphuric acid. The powder is well mixed with the acid and allowed to stand overnight at 20° to 22° C. The contents of the beaker are then made up to 800 ml. with water and boiled for 2 hours, the volume being kept constant by the frequent addition of water. The suspension is filtered through a Gooch crucible with a No. 42 Whatman filter paper, and the precipitate is dried, weighed, and then ignited, the lignin content being calculated on an ash-free basis. F. A. R.

Inorganic

Determination of Arsenic in Organic and Inorganic Compounds. D. T. Lewis and V. E. Davis. (*J. Chem. Soc.*, 1939, 284-286.)—For the gravimetric determination of arsenic the authors favour its precipitation as uranyl ammonium arsenate, a slimy gelatinous precipitate quite insoluble in acetic acid and convertible by ignition into uranosic oxide U_3O_8 . The arsenate solution is treated with 10 ml. of 4 N ammonia, and acetic acid

until a faint odour is perceptible; it is boiled, and an excess of uranyl acetate solution is added. The precipitate, which becomes coarser after some hours' standing, is collected, washed, and ignited, leaving a moss-green residue of slightly reduced oxide. This is dissolved in a little strong nitric acid, which is cautiously evaporated; the final residue is ignited over a Bunsen burner, leaving U_3O_8 , which is weighed. Arsenite is not readily oxidised to arsenate by nitric acid, but dropwise addition of potassium bromate solution at 70° C. effects rapid oxidation. For the simultaneous determination of arsenite and arsenate, the arsenite in the solution is first titrated with bromate, after which the total arsenate is determined by precipitation with uranyl acetate, arsenate being computed by difference. Organic arsenicals may be oxidised by the Carius method, but some decompose explosively in contact with fuming nitric acid. A safe and efficient method consists in introducing 0.1 to 0.3 g. of the substance into a Kjeldahl flask containing a cold mixture of 20 ml. of strong sulphuric acid, 10 g. of potassium nitrate and 0.25 g. of pure starch. After gentle warming the heat is gradually increased, and the acid is boiled until all of the organic matter has been oxidised. After cooling, 40 ml. of cold water are cautiously added, followed by bromate solution until the liquid is coloured by bromine. The solution is neutralised with strong ammonia, filtered, slightly acidified with acetic acid and treated with uranyl acetate solution, and the determination is completed as before.

[It would seem that incorporation of filter-pulp with the uranyl ammonium arsenate precipitate would expedite filtration, washing, and the expulsion of arsenic by ignition.—ABSTRACTOR.] W. R. S.

Determination of Chromium in Chrometanned Leather. R. M. Lollar. (*J. Amer. Leather Chem. Assoc.*, 1940, **35**, 443-452.)—In the method described below, the ash is treated with perchloric acid, and the interference of ferric iron in the iodimetric titration of chromate is prevented by addition of phosphoric acid. The weighed sample (2 g.) is ignited in a nickel crucible, finally in a muffle. The ash is transferred to a conical 125-ml. flask, and gently boiled with perchloric acid of constant b.p. (about 71 per cent.) until the colour of the solution changes from green to orange. This may take from a few minutes to several hours. The acid solution, while still hot, is cautiously diluted with water and poured into a 400-ml. beaker, and the flask is rinsed with water (total volume, 150 to 200 ml.). The liquid is boiled for a short time to expel any chlorine, left to cool completely, and acidified with 25 to 30 ml. of phosphoric acid (1:1). After addition of 10 ml. of 10 per cent. sodium iodide solution the liquid is set aside for one minute out of the sunlight, and titrated with 0.1 N thiosulphate solution, starch being added towards the end. A rapid dilution of the hot perchloric acid is found necessary to prevent risk of formation

of hydrogen peroxide, which would reduce chromic acid. Potassium iodide in phosphoric acid solution gives a precipitate which masks the end-point; hence the sodium salt is used. The optimum concentration of phosphoric acid for the titration is 3 *N*; at higher concentrations ferric iron reacts upon the iodide, whilst insufficient phosphoric acid causes too slow a reduction of chromic acid by the iodide.

W. R. S.

Colour Test for Elemental Sulphur. H. Sommer. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 368-369.)—The material is extracted with hot pyridine, and the solution is filtered if necessary. One-tenth volume of 2 *N* sodium hydroxide solution is added and after brief shaking at room temperature the colour of the pyridine solution is noted at once. A light sky-blue colour is produced with 1 part of sulphur in 100,000 of pyridine; the colour is greenish-blue with 1 part in 10,000, dark olive-green with 1 part per 1000 and deep red-brown with a 1 per cent. solution. A range of blue colours is given by the use of saturated sodium bicarbonate solution instead of sodium hydroxide, when the mixture is boiled for a few seconds. The colour is unstable. Both crystalline and amorphous sulphur respond to the test. No colour is given by sodium sulphide or sodium thiosulphate. Elemental selenium or phosphorus produces no colour. Pyridine may be replaced by aniline or triethanolamine, but the sensitiveness is lower. Carbon disulphide inhibits the reaction and halogenated hydrocarbons, such as chloroform, produce purple colours with alkali-pyridine mixtures. The blue colour is probably due to colloidal sulphur. References to similar reactions are found in the earlier literature.

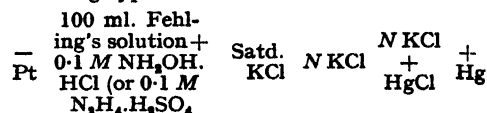
S. G. C.

Cerate Oxidimetry. Stability of Solutions. G. F. Smith and C. A. Getz. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 339-340.)—Quadrivalent cerium in nitric and perchloric acid solutions provides oxidation potentials of 1.6 and 1.7 volt respectively, whereas in molar sulphuric acid solution the potential is 1.44 volt. At the higher potentials the field of usefulness of the reagent may be extended, provided that the stability of the solution is not unduly sacrificed. Eighteen solutions of the nitrate and perchlorate cerate ions were prepared and examined. Nitric and perchloric acid solutions of hexanitrate ammonium cerate in nitric and perchloric acids between 1 and 3 *M* were the most promising. A 0.1 *N* solution of commercial hexanitrate ammonium cerate in 1 *N* perchloric acid was sufficiently stable in ordinary light not to require re-standardisation for a period of 20 days. The stability of all solutions improved on keeping, as the accumulation of cerous ions lowered the oxidation potential.

S. G. C.

Potentiometric Determination of Hydroxylamine and Hydrazine in Alkaline Solution. H. T. S. Britton and M. Königstein. (*J. Chem. Soc.*, 1940, 673.)—The

reaction of hydroxylamine and of hydrazine with both Fehling's solution and ammoniacal copper sulphate solution can be followed potentiometrically by using a system of the following type:



The complete reduction of the bivalent copper is indicated by a sudden reduction of potential. To prevent diffusion, the ends of the saturated potassium chloride bridge were plugged with asbestos and one end was immersed in a vessel containing *N* potassium chloride solution, in which was inserted also the tube leading from the normal calomel electrode. The titration vessel was fitted with an ebonite cap containing holes for two platinum electrodes, a thermometer, a tube leading from the burette, the end of the salt bridge, a mechanical stirrer, and a tube for the introduction of nitrogen. The whole cell was immersed in a water-bath kept at such a temperature that the solution in the titration vessel was at 90° to 92° C. Sodium tartrate was used instead of Rochelle salt in the Fehling's solution. When ammoniacal copper sulphate was used, ammonia was passed through the solution instead of nitrogen (*cf.* Britton and Phillips, *ANALYST*, 1940, 18, 149).

E. M. P.

Detection of Thiocyanate, Iodide, Bromide and Chloride. D. Hart and R. Meyrowitz. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 318-320.)—Tests on a separate portion of the solution are made for each anion, without the use of silver as a group reagent. Provision is made against the presence of the following ions: ferricyanide, ferrocyanide, cyanide, sulphide, thiosulphate, arsenite and tartrate. **Thiocyanate.**—Large amounts of iodide interfere with the detection of thiocyanate by means of ferric iron. Addition of lead nitrate to the solution acidified with nitric acid precipitates much of the iodide; ferricyanide is precipitated on addition of cobalt nitrate; thiocyanate is tested for in the filtrate by means of ferric iron. **Iodide** is detected by shaking the solution with carbon tetrachloride and hydrogen peroxide after removal of interfering substances as follows:—Ferricyanide, ferrocyanide, sulphide and cyanide are precipitated from the solution, slightly acid with sulphuric acid, by the addition of cobalt acetate (in absence of the first two ions, cyanide and sulphide may be removed by simply boiling the acidified solution); arsenite is precipitated with hydrogen sulphide; thiocyanate is destroyed by boiling for 30 seconds in 6 *N* sulphuric acid. **Bromide** is identified by shaking the solution with successive portions of carbon tetrachloride and a little potassium permanganate until the aqueous layer remains pink; a yellow or brown colour in the carbon tetrachloride layer indicates bromide. Interfering ions are first

removed. Thiosulphate is decomposed by boiling the acidified solution; mercuric acetate is added, followed by cobalt acetate to precipitate ferricyanide; any tartrate is precipitated with lead acetate. The precipitate is filtered off, mercury in the filtrate is precipitated with hydrogen sulphide and the liquid is filtered again. Thiocyanate is decomposed as in the test for iodide, and iodide is removed by boiling with sodium nitrite in dilute sulphuric acid solution. *Chloride*.—Ferricyanide, ferrocyanide, cyanide and sulphide are removed as indicated above. Iodide, bromide, thiocyanate and thiosulphate are removed by steam distilling the solution containing 30 per cent. of conc. nitric acid. A small amount of bromide remains, but this is removed by shaking the solution with carbon tetrachloride and potassium permanganate; the colour of the excess permanganate is subsequently destroyed with sodium nitrite, and any colloidal sulphur remaining from the decomposition of thiosulphate is removed by shaking with black mercuric sulphide and filtering the liquid. Chloride is detected with silver nitrate.

S. G. C.

Microchemical

Micro-gravimetric Determination of Active Hydrogen by the Grignard Reagent. R. N. Evans, J. E. Davenport and A. J. Revukas. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 301.)—A micro-gravimetric Zerewitinoff method is described in which the evolved methane is burned to carbon dioxide and water, which are absorbed in microchemical absorption tubes and weighed. The method is applied to the analysis of impregnated paper insulating tapes. The second hydrogen in the water molecule is active toward the Grignard reagent. Preliminary tests with the method show that theoretical results may be obtained for the active hydrogen content of typical organic compounds at 25°C. in absence of certain types of peroxides. The apparatus is the same as that employed by the authors in the determination of water in impregnated paper insulating tapes (*Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 553) with slight variations, in that the reagent is introduced into the reaction chamber through a serum rubber stopper, commonly employed in medical laboratories, by means of a syringe fitted with a stainless steel needle, and that two reaction cells in series are used, so that the reagent siphons on to the sample without air contamination. The rate of gas flow is approximately 1 litre per hour for the combined gases at the exit end of the train.

J. W. M.

Micro-determination of Potassium. I. A. Kaye. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 310–311.)—A variation of the cobaltinitrite method, using ceric sulphate instead of potassium permanganate in the titration, is applied to the determination of amounts of potassium ranging from 0.0200 to 0.1200 mg. About 0.5 ml. of the test solution is placed in a 15-ml.

centrifuge tube, 0.5 ml. of sodium cobaltinitrite reagent (Kramer and Tisdall, *J. Biol. Chem.*, 1921, 46, 339) is added dropwise with shaking, and the contents of the tube are left for 1 hour at 20°–25°C. The precipitate is centrifuged for 10 minutes at about 2000 r.p.m., the supernatant liquid is carefully removed and 5 ml. of water are added so as not to disturb the precipitate. Centrifuging and washing are carried out 3 times in all.

Reagent.—About 4.5 g. of anhydrous ceric sulphate are dissolved in 500 ml. of water to which 100 ml. of conc. sulphuric acid have been added. The solution is diluted to 1 litre and standardised with standard sodium thiosulphate. One ml. of this reagent is added for amounts of potassium less than 0.06 mg., the tube is heated in a water-bath for 2 or 3 minutes, until the precipitate dissolves. The solution is cooled to room temperature, 1 drop of 1 per cent. potassium iodide solution is added, and the liberated iodine is titrated with standard (0.002 N) sodium thiosulphate, a few drops of 0.2 per cent. starch solution being added near the end-point. At the same time 1 ml. of the ceric sulphate solution is titrated with the sodium thiosulphate.

J. W. M.

Identification of Perchlorate, Persulphate and certain other Inorganic Acid Radicals with Zwikker's Reagent. A Sensitive Reaction for Copper. G. H. Wagenaar. (*Pharm. Weekblad*, 1940, 77, 465–468.)—Zwikker's reagent (a mixture of 4 ml. of 10 per cent. copper sulphate solution, 1 ml. of pyridine and 5 ml. of water) will detect barbituric acid derivatives, saccharin and organic acids (*cf. id.*, 1931, 68, 975; 1932, 69, 1186; and *ANALYST*, 1934, 59, 850). In tests with 31 acid radicals amorphous precipitates were obtained with iodides, cyanides, thiocyanates and molybdates. Perchlorates gave light purple to blue, square and hexagonal crystals, which coalesced as they grew (sensitiveness, 1:100); scratching was unnecessary, and chlorates did not interfere. The crystals from persulphates were blue, very regular right-angled prisms, which coalesced into rectangular groups if the solution was strong (sensitiveness, 1:1000); some small black squares were also observed; scratching helped in this instance. Thiosulphates (1:1500) formed groups of long radiating needles, which were unaffected by the presence of sulphites. Chromates (1:300) crystallised in yellow X-shaped groups of rectangular plates, and there were also some isolated narrow rectangles which usually had a rectangular indentation in each of the narrow sides, and sometimes in all sides, giving an H-shaped appearance. Dichromates gave amorphous or microcrystalline precipitates. The shape of the crystals obtained from permanganates was dependent on the concentration of the solution; they were aggregates either of square plates or of dark needles (1:3000). This last reaction also affords a sensitive test for copper, 1 drop each of pyridine water (1:10) and N potassium permanganate solution being added to the solution

under examination, when plates of irregular shape or small needles are formed (sensitivity, 1:25,000 of Cu⁺⁺). Certain organic substances also react, e.g. when a small crystal of sodium sozoiodolate is added to a drop of the reagent a brown precipitate, which forms star-shaped groups of fine crystals, is produced. Illustrations are given of the various forms of crystals. J. G.

Micro-titration of Selenium. G. Wernimont and F. J. Hopkinson. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 308.)—A simple and accurate volumetric method, using the dead-stop end-point electrometric titration (Pring and Spencer, *ANALYST*, 1930, 55, 375; *et al.*) is applied to the determination of minute amounts of selenium, e.g. in urine. The precision is claimed to be $\pm 2\gamma$ in the range 2 to 150 γ on a basis of a 100-ml. sample; the method is sensitive to 0.02 p.p.m. *Detail.*—To 100-ml. samples of urine (containing abnormal amounts of selenium) 25 ml. of conc. sulphuric acid are added, in 300-ml. Kjehldahl flasks, together with 0.7 g. of mercuric oxide and several glass beads. The flasks are heated until the water has evaporated and the organic matter has been destroyed (about 3 hours). The clear solutions are transferred to 300-ml. flat-bottomed flasks, 60 ml. of water being used for rinsing. Into each flask are introduced 25 ml. of conc. sulphuric acid, 50 ml. of 48 per cent. hydrobromic acid containing 1 per cent. (by vol.) of liquid bromine, and the ground joint is moistened with conc. sulphuric acid. The flasks are then attached to small Liebig condensers and the mixtures are distilled until 35 to 40 ml. of distillate have been collected. The distillate is treated with 5 per cent. sodium sulphite solution and then with 5 per cent. phenol solution to destroy the bromine. Standard (0.001 N) sodium thiosulphate solution is then added to give an excess of 2 to 3 ml. After addition of 1 ml. of freshly prepared 5 per cent. potassium iodide solution, the excess of sodium thiosulphate is immediately titrated with 0.001 N potassium iodate solution, the dead-stop method being used to determine the end-point. J. W. M.

Physical Methods, Apparatus, etc.

Machine for Testing the Fastness to Rubbing of Dyed Materials. D. A. Derrett-Smith and H. B. Bradley. (*J. Soc. Dyers and Colourists*, 1940, 56, 261–265.)—The fastness to rubbing of a dyed material is defined as the extent to which it gives rise to a coloured mark when rubbed against a white material, and it is best determined by means of a machine which rubs under standardised conditions. Existing methods (*cf.* German Fastness Commission, "*Verfahren, Normen und Typen*," Verlag Chemie, Berlin, 1935; Forster, Ramachandran and Venkatamaran, *J. Soc.*

Dyers and Colourists, 1938, 54, 216) are discussed, and it is concluded that there is a need for a simple machine which produces a rubbing effect as near as possible to that produced by hand tests, under standard conditions and in a comparatively short time. In the machine now described a strip of plain weave bleached dress linen is held between a fixed and a detachable clamp over a flat aluminium platform, the detachable clamp being connected with the overhanging end of the cloth which is kept taut by means of a weight attached to this clamp; the weight and clamp together weigh 820 g. The dyed material to be tested is wrapped round a hard beech-wood block which is attached to a light metal carriage by means of screws and wing-nuts. The block also carries a detachable weight which ensures close contact between it and a small area of the dyed material. A heart-shaped cam driven by a geared electric motor and attached to the block-carriage by means of connecting rods, causes the block carrying the dyed material to travel 5.5 inches backwards and forwards along the bleached cloth, at the rate of 84 rubs per minute. Numerous tests on materials dyed with azo-colours are described, these being chosen because one of the factors governing their fastness is believed to be substantivity of the particular Brenthol used; the lower the latter, the greater the proportion of dye held mechanically in the final dyeing (*cf.* Blackshaw, *id.*, 1936, 52, 135). It was established that with dyeings of relatively high fastness it is best to compare the development of the coloured mark on the bleached material after a definite number of rubs (e.g. 500) with each of the dyed materials to be tested. When the fastness is low (e.g. when a mark is produced after 50 rubs) the test is carried out in 3 stages, each of 20 rubs, the bleached cloth being moved along in the clamps in such a way that a separate record of the effect of each stage is obtained on the same sample; in this way it is possible to ascertain whether the dye is being rubbed off on to the bleached material at a uniform rate, or otherwise. Alternatively, a particular dyeing may be taken as a standard, measurements being made of the number of rubs necessary to produce the same depth of marking as is obtained from a given number of rubs on the standard. Actual numerical measurements of the coloured marks may be made with the Lovibond Tintometer. Damping enhanced the severity of the test, more colour being rubbed off. Yarn dyeings from Brenthol-BN were found to be faster than those from Brenthol-AS or -OT, which were similar in this respect; with piece dyeings a decreasing order of fastness was obtained from Brenthol-BN, -CT and -AS. Vat dyeings on linen were comparatively fast when dry, 200 rubs giving no discernible mark, although yarn dyed with indigo gave a faint mark after 100 rubs. J. G.

Luximeter: Construction and Application. W. L. Carson. (*Gen. Elec. Rev.*, 1940, 43, 91–92; *J. Text. Inst.*, 1940, 31, A 341.)—The

Luximeter is a simple photo-electric instrument for the measurement of the transmission of light through liquids. It was designed originally for use in the phosphatase test for the pasteurisation of milk, but has many other applications in chemical analysis and industrial process control. Light from a 6- to 8-volt Mazda lamp, which can be maintained at a constant intensity, passes through a test-tube containing the sample to the photo-electric cell, which actuates a sensitive indicating instrument. The intensity of light required for a given test varies according to the opacity of the sample, and is controlled by means of a resistance in the lamp circuit. Readings taken on a light meter, which is similar to the common exposure-meter except that it is divided arbitrarily into 100 divisions, are plotted against the concentrations for solutions of known strength, and this curve is used to obtain the concentrations of unknown solutions from the corresponding readings on the light meter.

J. G.

Optical Method for the Determination of Quartz Particles in Felspar. G. H. McIntyre and M. Bozain. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 326-328.)—A 25-g. sample of the finely powdered felspar is mixed with sufficient gum arabic solution to make a stiff paste. This is dried in a mould and the cake is subsequently fused at a temperature gradually rising to 2500° F. to yield a translucent glass, in which the quartz particles remain suspended. The cooled button is pulverised to pass a 200-mesh sieve. A small sample is mixed with a drop of glycerin and pressed between glass microscope slides, and the number of quartz particles per unit area of field is counted in polarised light. Standards are prepared by a similar technique from felspar of low quartz-content intimately incorporated by grinding with known proportions of quartz. A curve is plotted of the number of particles per unit area of field in relation to the percentage of free quartz, and from this the proportion present in the test sample may be determined.

S. G. C.

Reviews

CHEMISTRY IN THE SERVICE OF MAN. By ALEXANDER FINDLAY. Fifth Edition. Pp. xx + 398. London, New York, Toronto: Longmans, Green & Co., Ltd. 1939. Price 8s. 6d. net.

This book, in its fifth edition and its thirty-fourth year, seems to me to exercise the same fascination as I recall from the now so remote-seeming past when, as a schoolboy, I first devoured the contents of its revealing pages. Then, as to-day, if less consciously, I was carried away by the author's enthusiasm for his subject, pride in his colleagues and patience with his readers. A perusal of the third edition, nearly ten years later, impressed one even more with the author's courage. Writing a book of this kind, intended primarily for the non-practitioner, if not for the layman, is rather like teaching an illiterate to read. There are two possible lines of approach. He can gradually be trained up to a point where he is familiar with all the words of one syllable, after having been taught his alphabet, by appropriate use of simple pictures and monosyllables. But there his education will end, for he has no grammar, no understanding of the structure of sentences and no possibility of understanding the subtler uses of language. The other method is to teach literacy itself, with all that this implies about knowledge of words and sentences and the logic of the written medium. Professor Findlay's method is analogous with the second, and in this respect differs from that of less courageous or less energetic writers. He does not hesitate to take his non-chemical reader—and to take him at the very beginning of the book—into discussions on radio-activity and atomic structure. By the end of the third chapter the phlogiston theory has been met and disposed of. And so it goes on, the reader being led rapidly, but with logic and clarity, through the wide ramifications of modern chemical science, with head in the exciting empyrean of theory, feet on the solid earth of practice.

Professor Findlay of all men clearly believes that accurate statement of facts justifies any amount of careful explanation, for it is the groundwork of the reader's understanding that he is attempting to build, and this itself must never be liable to disturbance through the existence and subsequent detection of faulty units of construction. He will, therefore, doubtless forgive a reviewer for calling attention to one or two minutiae. Modern commercial casein (p. 367) is certainly never

prepared by solution in ammonia and subsequent precipitation, but by careful control of the "starter" (in the self-souring process) or of the pH produced by the addition of hydrochloric acid (in the grain-curd process). It would have been well to point out that rennet casein differs chemically, as well as physically, from the acid casein used in the paper-making and paint industries. The food use of casein is so trivial that it is hardly worth mentioning.

Again, it is not correct to say, nor was it at the time Professor Findlay passed the proofs, that vitamin B₁ "is not yet an industrial product." Synthetic aneurin has been on the market for at least two years, in this country as well as in Switzerland, Germany and the United States. Finally, synthetic ascorbic acid, being made from optically active sorbose, is not racemic, but identical with the natural product.

But these are, as I have admitted, *trivia*, and only mentioned in the interests of the accuracy that Professor Findlay so clearly holds dear. The book remains what it has always been—a masterpiece of lucid and enthralling exposition. Every chemist should see to it that his children have this book. There is no doubt whatever that they will read it, and little less doubt that their father would be well advised to read it also, so that he may meet the cross-examination that is sure to follow.

A. L. BACHARACH

COLLOID CHEMISTRY (A TEXTBOOK). By H. B. WEISER. Pp. viii + 428. New York: John Wiley & Sons, Inc.; London: Chapman & Hall, Ltd. 1939. Price 24s. net.

Students of elementary colloid science are now well provided with textbooks from the pens of the recognised masters, and surely with the appearance of Professor Weiser's volume the bookshelf is complete. If one admits that there was room for another introductory text, then praise must be accorded to this most recent effort.

To quote the Preface: "The plan of the book is as follows: After an introductory chapter, which is concerned with the general aspects of the colloidal state of matter, appears a section of seven chapters dealing with the phenomena of adsorption at various types of interfaces. Following this fundamental section on adsorption, ten chapters are devoted to the formation and properties of lyophobic and lyophilic sols. This discussion is followed in turn by shorter sections on gels, emulsions and foams, and aerosols and solid sols. The three concluding chapters deal in a more comprehensive way with the application of colloid chemical principles to contact catalysis, dyeing and clay. An attempt has been made to render the presentation as clear and concise as possible by outlining the subject matter with frequent section and paragraph headings."

When the author, himself a notable investigator, tells us that various sections have been approved by Abramson, Bancroft, Briggs, Bradfield, Bartell, Bull, Foulk, France, Garrison, Milligan, Harkins, Holmes, McBain, Miller, and Williams, a challenge faces any reviewer. These names spell authority. Deeper interest is added to the usual pleasurable reading of such volumes.

The present reviewer unreservedly recommends this textbook as a very good introduction to colloid chemistry, written in agreeable style. Modern views are well treated on the whole, although space has not permitted detailed discussion on certain points deserving of fuller treatment, such as emulsions, hydrophilic sols, and, in particular, colloidal electrolytes. Adsorption phenomena are well treated in 113 pages, whilst sols occupy 149 pages.

Printing and binding leave nothing to be desired.

WILLIAM CLAYTON

INTRODUCTORY COLLEGE CHEMISTRY. NEIL GORDON and WILLIAM E. TROUT, JR.
Second Edition. Pp. ix + 753. New York: John Wiley & Sons, Inc.;
London: Chapman & Hall, Ltd. 1940. Price 21s.

Gordon and Trout's introductory textbook, now in its second edition, opens in a very elementary way with the chemistry of water and leads on, as far as possible by experiment, through the chemistry of the usual non-metallic elements (oxygen, hydrogen and nitrogen). On the way such subjects as molecular and atomic weights are dealt with. The electrical theory of matter is introduced at a very early stage and is used freely to explain various facts of chemistry. Theories of ionisation, including that of Brönsted, are discussed in considerable detail. After sections on sulphur and the halogens the Periodic Classification is treated fully and explained in the light of modern ideas of atomic structure. An account of carbon, phosphorus, the metalloids and the colloidal state completes the first half of the book.

In dealing with the metals the periodic classification is, surprisingly enough, discarded and the subject is treated from the point of view of the groupings in the common group analysis tables. This unorthodox approach is justified on the grounds that it permits of an experimental treatment. The book closes with a brief account of certain periodic groups, including those of the rare earths and the inert gases.

One of the most striking features is the apparent chaotic medley of methods used for presenting the facts. A closer examination, however, reveals very sound reasons for this. Introductory College Chemistry is what its title suggests—an introductory textbook—and the authors have tried to introduce the subject experimentally, wherever possible, but, where the nature of the subject demands it, have adopted the usual didactic method of statement. Thus, interposed between experiments, which must be performed by the student, unfinished equations, blank tables and unanswered questions, one finds very good accounts of such subjects as the law of mass action, ionisation, and atomic numbers. The book is naturally very elementary but, if rightly used, should form a satisfactory introductory course. It is well bound, free from errors, and altogether very attractive.

HAROLD TOMS

KINGZETT'S CHEMICAL ENCYCLOPAEDIA. Revised and Edited by RALPH K. STRONG, Ph.D., with a Foreword by Sir GILBERT T. MORGAN, O.B.E., F.R.S., D.Sc., LL.D. Sixth Edition. Pp. x + 1088. London: Baillière, Tindall & Cox. 1940. Price 45s.

"At a time of national emergency a chemical encyclopaedia becomes more than ever essential, for it gives a concise introduction to the most fundamental of all industries, the chemical trades and manufactures which subserve the community's needs for food and clothing, shelter and defence." So wrote the late Sir Gilbert Morgan in his foreword to this edition of "*Kingzett's Chemical Encyclopaedia*." This is surely sufficient justification for the publication of such a book at one of the most critical times in our national history, when, possibly for the first time, innumerable non-technical administrators find themselves compelled to wrestle with the problems of a highly technical science and its applications to various industries. For them, as well as for chemists and others engaged in the chemical industry, this encyclopaedia will furnish most of the general information they are likely to need. The references appended to many of the sections will be found useful when more detailed information is required.

Unfortunately, Sir Gilbert Morgan, who had undertaken the task of revising the Encyclopaedia at Mr. Kingzett's request, found himself unable to do so owing to ill-health, and the revision was undertaken by Dr. R. K. Strong. Sir Gilbert died before the book was published, but he was able to assist Dr. Strong by reading the manuscript and proofs.

In essence, the new edition is identical with the original work, and its aim is still that laid down sixteen years ago by Kingzett, ". . . to prepare an epitomised digest of chemistry and its industrial applications, in a form which should be useful as a work of reference by all classes of the community." Probably no other one-volume encyclopaedia has achieved this so successfully as the book under review, for it covers every conceivable aspect of chemical theory and practice and the applications of chemistry in industry and commerce. Even the economic aspect is dealt with in this new edition, although it is doubtful how far statistics, already four years old, will be of permanent value.

He would be a bold reviewer—and an omniscient one—who would dare to pass judgment on this book *in toto*, and the present reviewer has been content to judge the book by its references to that section of chemistry with which he is most familiar. For the rest, he can only express his unbounded admiration for the enormous mass of data collected within the confines of its covers.

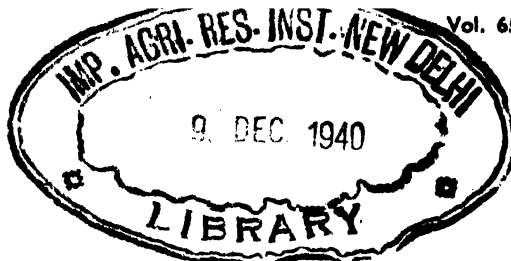
A few errors are to be expected in a work dealing with such a vast and widely-dispersed literature, and due allowance must be made if the accounts do not always include the results of the latest researches. Nevertheless, it is a little surprising to find under the heading "Sterols," that "There are many isomers, it is supposed, of cholesterol, but the available data respecting them, although of biological importance, is very vague"; such a statement was doubtless true in 1933, which is the last year referred to in the bibliography, but surely not in 1940. Similarly, the subject "Hormones" is most inadequately treated, the latest reference being to a paper published in 1931! Under the heading "Ergot," ergometrine, the most important alkaloid isolated, is not mentioned at all, yet it was isolated in 1934. On the other hand, some of the sections, such as those on "Vitamins" and "Legal Matters," are up to date and contain quite modern references. Some of the descriptions also suffer from a certain lack of precision. Thus, many would regard the phrase "used in medicine" as hardly a sufficiently precise description of the uses to which such substances as codeine and methyl salicylate are put. Similarly, the regular use of empirical formulae, where structural formulae might have been included without much trouble, is to be deplored.

The following are some of the more important errors noted. On p. 10, sulphanilic acid is stated to be the 1:3 isomer. "Corpus luteum" is described as a hormone on p. 259. "Oestrone" does not appear at all, but is described under the older designation "Theelin," which is no longer used. "Lachrymator" is spelt "lacrimator." Finally, it is stated under "Anaesthetics," that the first person to use ether for this purpose was William Morton in 1846, whereas the discovery is usually attributed to Crawford Long, who used it in 1842.

There are also certain rather surprising omissions. Thus, under "Hydrogen Ion," a comprehensive description is given of the use of the hydrogen electrode, but the quinhydrone and glass electrodes are mentioned only in the bibliography. Again, sulphanilamide is included, but not prontosil, or any of the other members of the sulphonamide group which are nowadays of such great importance.

It will be realised, of course, that the subject matter of these criticisms is a mere fraction of the mass of sound information which constitutes the bulk of the book. The true test of the usefulness of such an encyclopaedia as this is the value which all chemists in every branch of the science place upon it, and there can be no doubt at all about the warm reception that this new edition will receive.

F. A. ROBINSON



THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

THE following candidates have been elected members of the Society:

James Norman Davidson, B.Sc., M.D. (Edin.), University Lecturer on Biochemistry. (*Through the Scottish Section.*)

James Ross Fraser, B.Sc. (Lond.), F.I.C., Chemist in Government Department.

Granville Nicholas Gee, Chemist and Dyeing Technologist.

Daniel Evans Jones, M.Sc. (Wales), F.I.C., Chemist in Public Health Laboratory.

Francis Laurence Kinsella, Chemist with Feeding Stuffs Manufacturers. (*Through the North of England Section.*)

Robert Karran Matthews, F.I.C., Chief Assistant to Public Analyst and University Lecturer on Chemistry. (*Through the North of England Section.*)

William John Puregger, Branch Manager and Chief Chemist to Public Analysts in Australia.

Robert Edwin Stuckey, B.Sc. (Lond.), A.I.C., Ph.C., Staff Chemist at Forensic Science Laboratory. (*Through the North of England Section.*)

John Adam Cunningham Watson, A.I.C., Chemist with Feeding Stuffs Manufacturers. (*Through the North of England Section.*)

STATUTORY RULES AND ORDERS, 1940, No. 606

The Dripping (Maximum Prices) Order*

THE Society having been asked to suggest an appropriate fee for the analysis of samples of dripping submitted to Public Analysts by the Food Controller under this Order, the Emergency Committee of the Council has considered the matter and suggests that a suitable fee would be that recommended for the determination of meat in sausages in the circular letter sent to Public Analysts in May, 1940.

* cf. p. 567

The Composition of some Jam Fruits and the Determination of the Fruit Content of Jams

By C. L. HINTON, F.I.C., AND T. MACARA, F.I.C.

(Read at the Meeting, May 1, 1940)

IN an earlier paper¹ analytical data for the proximate composition of samples of a number of jam fruits were summarised. It was pointed out that there could be no finality in the figures, since the districts of supply, the varieties in cultivation, and the effects of seasonal conditions would all be liable to change over a period and thereby affect the average figures or the range of extremes. With a view to keeping in touch with any such trends, and to extending the data in some directions, further samples of fruits have been examined in the laboratories of the Research Association as opportunity permitted, and the results are given in the present paper.

Many of the samples were purchased locally, some were obtained direct from Covent Garden Market and others were samples from deliveries of jam fruit used by members of the Research Association. Included are a number of samples of fruit grown on the Continent.

The present data include figures for certain analytical characteristics not investigated in the earlier work, *viz.* combined acid (and hence also total acid), lead numbers and *pH*. These additional figures are the result of the working out of the lead precipitation method,² and the fact that experience over a number of years has shown this method to have considerable value in the examination of jams. Some of these figures were summarised in the paper mentioned,² but they are now given in detail.

ANALYTICAL METHODS

A description is given of the methods used where these have not already been described,^{1,2} or where alterations in the procedure have been made.

Preliminary Preparation of Sample.—Portions of the well-mixed minced material were quickly weighed out in duplicate for: (a) Determination of insoluble solids, seeds and pectin; (b) Preparation of an aqueous extract on which the determinations of soluble solids, acidity, etc., could be made.

In preparing stone fruits the stones from a weighed quantity of fruit were scraped free from any adherent tissue, allowed to dry in the air and weighed. Only the fleshy part of the fruit was analysed, and the analytical data for these fruits refer to the flesh only.

Insoluble Solids.—The residue of fibre, etc., after the first boiling and filtration, was boiled with two further quantities of water for periods of 15 minutes each and finally washed thoroughly on the filter. Filter and contents were dried overnight at about 105° C. All the filtrates and washings were reserved for the determination of pectin (see later).

Seeds.—The dried insoluble matter was detached from the filter, and boiled for a short time in weak sodium hydroxide solution (about *N*/50), to open the material and clean the seeds. These were allowed to subside, the alkaline liquor was poured off, and the seeds and fibre were boiled with water and then transferred to a filter and washed with hot water. The residue was turned out on to a white tile, and the seeds were teased out with a spatula, placed in a weighed box, dried at 105° C., cooled and weighed.

Pectin.—Experimental work in these laboratories has shown that pectin-containing fruit tissues tend to give up their pectin in varying degree according to the nature of the liquid by which they are extracted. It has, of course, been known

for some time that the *pH* of the extracting liquid is important. It now appears that the nature and concentration of salts in fruit juices can also have a marked effect. When a fruit is boiled with water, the pectin is being extracted not by water only, but by a diluted solution of the juice of the fruit. The composition of this solution will vary according to the composition and dilution of the juice. It is thus clear that the conditions of extraction of the pectin when the fruit is simply "boiled with water" may vary considerably.

In particular, it may be noted that in the preparation of an aqueous extract of the fruit, the pectin is being extracted by a fairly concentrated solution of the juice. When, however, the fruit is boiled with a fair amount of water, and the fibre, etc., is then washed copiously with boiling water, the pectin is virtually being extracted by an excess of distilled water only. It appears that the amount of pectin extracted by the latter procedure may be appreciably greater than by the former. Table I shows some results obtained by the two methods of procedure. Smaller differences of the same kind have been observed repeatedly also in the analysis of jams. It appears that they may partly, but cannot entirely, be accounted for by adsorption effects.

TABLE I—AMOUNTS OF PECTIN EXTRACTED FROM FRUITS BY DIFFERENT METHODS

| Mode of extraction, etc. | Pectin extracted from | | |
|---|--|------------------------------------|------------------------------------|
| | Blackcurrants (canned) Per Cent. | Raspberries (pulp) Per Cent. | Raspberries (pulp) Per Cent. |
| Boiled with an equal weight of water for 1 hour, and filtered, but not washed through. Pectin determined in an aliquot part of filtrate | { (1) 0.39 (2) 0.35 | 0.13 | 0.08 |
| Boiled with successive portions of water and washed through well | { (1) 0.57 (2) 0.57 | (1) 0.33 (2) 0.27 | (1) 0.22 (2) 0.18 |

In view of this, it was thought desirable to modify the method for determining pectin in fruits, by applying it to the filtrate and washings from the insoluble matter. In some of the determinations the whole of the filtrate and washings was used, in others an aliquot portion. The subsequent procedure was that of the Carré-Haynes determination, without preliminary separation of the pectin with acetone; thus the results must be regarded as "crude calcium pectate."

Figures for pectin in fruits given in the earlier paper¹ are probably somewhat low in comparison with the present results, the pectin having been determined on aliquot parts of a 20 per cent. extract.

Soluble Solids.—For this and other determinations of soluble constituents, 250 g. of the minced sample were gently boiled with 250 ml. of distilled water for an hour, with frequent stirring. After cooling, the mixture was made up to 500 g., well mixed and filtered through paper or a fine sieve. The soluble solids content of the extract was then determined, usually by means of the specific gravity,³ but in some instances by both specific gravity and dipping refractometer, the results being averaged.

The figures so obtained were in g. of soluble solids per 100 ml. of the extract. In preparing the extract, 100 g. of fruit yield only 200—I g. of total liquid (where I = percentage of insoluble solids in the fruit). Moreover, this (200—I) g. of liquid occupy only $\frac{(200-I)}{D}$ ml. in volume (D being the density or, sufficiently closely, the sp.gr. of the extract). Hence, to convert the results to the percentage by weight in the original fruit, they were multiplied by $\frac{(200-I)}{100 D}$.

The same factor was applied, of course, to the results of other determinations carried out on the aqueous extract, *i.e.* acid and lead numbers.

Free and Combined Acid.—Previously to 1932 these determinations were made directly on the aqueous extract. Afterwards they were usually made on the pectin-free filtrate, by the procedure described earlier.² A few determinations were made on both the aqueous extract and the pectin-free filtrate; the differences, if any, were negligible.

All acidities, even those of malic acid fruits, are expressed for convenience as citric acid.

Lead Numbers.—These were determined as described in the earlier paper². In the analyses of the last few years, use has been made of an improved method for finding the end-point of the lead titrations, especially for the aqueous lead number. This is as follows:

A first rough titration is made, with the molybdate solution added in 0.5 ml. portions. A fresh portion of lead filtrate is then diluted, and molybdate solution is added to within about 0.5 ml. of the expected end-point (as shown by the rough titration). The mixture is boiled, the precipitate is allowed to settle, and the partly clear liquid is decanted through a fluted filter. The precipitate is then transferred to the filter and washed thoroughly two or three times with a jet of almost boiling water. Finally, the filtrate and washings are heated to boiling and the titration is completed, the molybdate now being added 0.1 or 0.2 ml. at a time, and the mixture spotted out with indicator after each addition.

It will be noticed that the figures shown for "lead number per 1 per cent. of total acid" correspond numerically to what was before expressed as "lead number per 0.1 g. of total acid."² This is so, of course, because the first expression refers to a unit total acid content in the sample of 1 per cent. and the second refers to a unit of total acid of 0.1 g. in 10 g. of sample on which the lead number is based, *i.e.* again 1 per cent. of the sample. The former mode of expression is rather more convenient, and in the present paper replaces the earlier one.

pH Value.—This was determined on the aqueous extract by means of the quinhydrone electrode in conjunction with a saturated calomel electrode. The formula for calculation of the pH from the reading in millivolts was: $\text{pH} = (454 - \text{mv.}) / 0.0001984T$, where mv. is the reading in millivolts, and T the absolute temperature as °C. As a control for the accuracy of the system, a standard buffer solution (0.4 per cent. solution of cream of tartar) was used, having a pH of 3.57.

TABLE II—pH VALUES OF GOOSEBERRY EXTRACTS DETERMINED WITH THE HYDROGEN AND QUINHYDRONE ELECTRODES

| No. | pH with hydrogen electrode | pH with quinhydrone electrode | |
|-----|----------------------------|-------------------------------|---------------------|
| | | Initial reading* | Subsequent readings |
| 1 | 3.00 | 3.04 | 3.13 (18 mins.) |
| 2 | 3.32 | 3.33 | 3.38 (7 ") |
| 3 | 3.04 | 3.09 | 3.13 (9 ") |
| 4 | 2.95 | 2.90 | 2.93 (5 ") |
| 5 | 2.90 | 2.89 | 2.91 (5 ") |
| 6 | 3.22 | 3.21 | 3.24 (6 ") |
| 7 | 3.21 | 3.19 | 3.24 (10 ") |
| | | | 3.40 (15 hours) |

* About 1 minute after mixing in the quinhydrone, except for No. 3 (3 minutes).

With some extracts, especially of gooseberries and currants, a marked drift of the apparent pH occurred. This drift sometimes amounted to as much as 0.1 unit in a few minutes, but varied considerably from one sample to another.

Some comparative determinations on gooseberry extracts showed that the initial value obtained with quinhydrone was usually quite close to that obtained with the hydrogen electrode. (Table II)

In determining the pH of the fruit extracts, therefore, the initial reading was taken to give as nearly as possible the correct value.

It seems possible that the drift is due to some action of reducing substances in the fruit juice on the equimolecular mixture of hydroquinone and quinone which is formed when quinhidrone dissolves. The principal substance likely to have a sufficiently powerful reducing action is ascorbic acid; but without further work it would not be safe to ascribe the effect definitely to this substance.

DETAILED ANALYTICAL DATA FOR THE FRUITS

It has been thought desirable to give the detailed analyses of each individual sample (Tables III to XV). This is the most satisfactory way of making them available for use in the interpretation of the analysis of jams, etc.; extreme and average figures have only limited value, since a sample which is extreme in one respect is usually more normal in other ways. An asterisk is placed against figures which are more extreme than any of the corresponding figures of the earlier paper.¹

Gaps in the tables occur because many of the samples were obtained primarily for other work, and time permitted the determination only of the analytical figures required for that work.

NOTES.—*Gooseberries*.—The averages, where comparison is possible, are not very different from those reported earlier,¹ but rather low minima have been found for soluble solids and free acid.

The aqueous lead number (per 1 per cent. of acid) is a measure of the approximate proportions of citric and malic acids in the fruit. If the fruit acids were entirely citric acid, the figure would be 13.55; if they were entirely malic acid, it would be approximately 5.8.² The average value of 10.36 found, corresponds to a mixture of 41 per cent. of malic and 59 per cent. of citric acid. The extremes, however (11.7 and 9.0) correspond to 24 per cent. and 59 per cent. of malic acid respectively.

Strawberries.—When the "plugs" had not been removed in picking, they were taken out as far as possible prior to analysis. The averages were close to the earlier ones,¹ and none of the extremes lay outside the ranges previously established.

The weight of seeds is a fairly constant proportion of the total insoluble matter, averaging 54 per cent., the extremes being 44 per cent. and 61 per cent.

The lead number (aqueous) per 1 per cent. of acid indicates an average proportion of about 11 per cent. of malic acid in the acids of this fruit. A range of variation from 0 to 40 per cent. was shown by individual samples. Only one sample approached the latter figure, however, and this was in the period before the introduction of the improvement in the titration mentioned above. No subsequent sample has shown a larger proportion of malic acid than 18 per cent.

There is a fairly definite relation between the pH and the proportion of combined to total acid. This can be seen from Fig. 1. The dotted line is the neutralisation curve for citric acid in $N/10$ concentration, and the points for the strawberry samples lie fairly near this line. The deviations from the citric acid curve are doubtless due to various causes, e.g. the fact that the neutralising bases are not entirely alkalies, but to some extent alkaline earths; the presence of small amounts of malic acid; variations in concentration from the decinormal; lack of accuracy in the various determinations involved. On the whole, in spite of these sources of deviation, the correspondence is quite good. It is obvious that the pH of a fruit extract is not an independent property, but is governed by the proportions present of the organic acids and their salts.

Raspberries.—For the purpose of arriving at the average and extreme figures of Table V the data for 24 samples, mostly of Scottish fruits, specially examined in 1934 and recorded previously (T. Macara),⁴ have been brought in.

In comparison with earlier figures,¹ the averages for soluble solids and free acid are about the same, but the average for insoluble solids is 1 per cent. lower. This must probably be attributed to the large proportion of samples of the Lloyd George

variety; this has come into considerable favour for jam-making, and it tends to have a smaller proportion of insoluble matter than other varieties. Its content of soluble solids and acid would appear to be more normal.

The weight of seeds averages 77 per cent. of the total insoluble solids, with a range from 70 to 82 per cent.

From the lead number per 1 per cent. of acid the average proportion of malic acid in the total acids is not more than about 3 per cent. In an extreme instance it was 19 per cent., but this sample was analysed in the early period of the lead precipitation method, and the result may be open to some doubt. Later figures, though few, indicate practically no malic acid in this fruit.

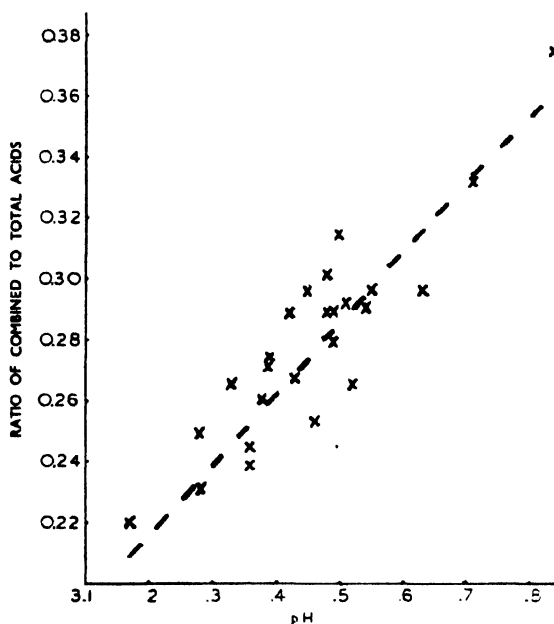


FIG. 1. RELATION BETWEEN pH OF STRAWBERRY EXTRACTS AND THE RATIO OF COMBINED TO TOTAL ACIDS.

[The broken line shown is the neutralization curve for citric acid.]

The relation between the pH and the proportion of combined acid in the total acid, as for strawberries, corresponds approximately to a partly neutralised solution of citric acid. Figures for pectin are not given, as it rapidly disappears from this fruit.

Redcurrants.—The averages on the whole are very close to the earlier ones.

The lead numbers, both aqueous and acetone, tend to be higher than can be accounted for by assuming the total acid present to be citric acid. (This would require values of 13.55 and 15.0 respectively for the lead numbers per 1 per cent. of total acid.) This seems to be due to precipitation of a certain amount of lead by the colouring matter of the fruit which, however, does not contribute to the acidity as determined by titration. The extracts from two samples were submitted to a decolorising treatment with vegetable carbon, and the lead numbers and total acid were determined on the decolorised solutions. It was found that the lead numbers per 1 per cent. of acid had fallen slightly, the acetone lead numbers now being close to the figure of 15.0 which would be expected from citric acid, or a mixture of citric and malic acids. (Table XVI)

TABLE III—GOOSEBERRIES

| Date | Description | Insol. solids Per Cent. | Sol. solids Per Cent. | Acid as citric (cryst.) | | | Lead numbers | | Lead number per 1% total acid | | Pectin Per Cent. | pH (50% extract) |
|------------------|---|-------------------------------|-----------------------------|-------------------------|--------------------------|-----------------------|--------------|---------|----------------------------------|---------|------------------------|------------------------|
| | | | | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Aqueous | Acetone | Aqueous | Acetone | | |
| 1930, June 2 | Small, green .. | .. | .. | 1.50 | 0.33 | 1.83 | 17.3 | — | 9.5 | — | — | 3.20 |
| " 4 | Green, hard .. | .. | .. | 1.99 | 0.32 | 2.31 | 23.8 | — | 10.3 | — | 0.99 | 3.14 |
| " 11 | Some partly ripe, not fresh .. | .. | .. | 1.91 | 0.36 | 2.27 | 24.3 | — | 10.7 | — | 0.76 | 3.30 |
| " 12 | Very small, partly ripe, not fresh .. | .. | .. | 6.2 | 1.70 | 2.09 | 24.5 | — | 11.7 | — | 0.59 | 3.07 |
| " 13 | Green .. | .. | .. | 7.5 | 2.06 | 0.37 | 23.4 | — | 9.6 | — | 0.86 | 3.37 |
| " 16 | Green, fairly fresh .. | .. | .. | 7.7 | 1.99 | 0.40 | 2.39 | — | 10.8 | — | 0.87 | 3.23 |
| " 25 | Firm, some beginning to ripen .. | .. | .. | 7.5 | 2.14 | 0.33 | 2.47 | — | 10.6 | — | 0.69 | 3.01 |
| " 26 | Large, green, beginning to soften .. | .. | .. | 10.2 | 2.49 | 0.33 | 2.82 | — | 10.6 | — | 0.79 | 2.84 |
| " 27 | Large, green, firm .. | .. | .. | 2.16 | 0.39 | 2.55 | 24.6 | — | 9.6 | — | 1.50 | 3.26 |
| " 30 | Large, green, some beginning to ripen, not very fresh .. | .. | .. | 2.26 | 0.35 | 2.61 | 27.3 | — | 10.5 | — | 0.82 | 2.90 |
| " July 3 | Green, but beginning to ripen, not very fresh .. | .. | .. | 7.7 | 2.04 | 0.34 | 2.38 | — | 10.1 | — | 0.64 | 3.02 |
| " 3 | Green and firm .. | .. | .. | 8.2 | 2.19 | 0.32 | 2.51 | — | 10.3 | — | 0.70 | 2.92 |
| " 17 | Green, but softening .. | .. | .. | 1.94 | 0.32 | 2.26 | 22.2 | — | 9.8 | — | — | 2.92 |
| 1931, June 15 | Medium to small, firm, moderately ripe .. | 2.48 | 7.8 | 2.24 | 0.36 | 2.60 | 28.6 | — | 11.0 | — | — | 2.96 |
| " July 1 | Medium to small, green, firm .. | 3.49 | 7.4 | 2.30 | 0.36 | 2.66 | 28.7 | — | 10.8 | — | — | 2.92 |
| 1932, June 1 | Mostly very small, green, firm .. | 2.25 | 6.5 | 1.27 | 0.38 | 1.65 | — | — | — | — | — | 3.40 |
| " 21 | Rather small, a few turning red .. | 2.24 | 8.2 | 2.26 | 0.39 | 2.65 | 28.1 | 40.9 | 10.6 | 15.4 | — | 3.01 |
| " July 11 | Green and hard .. | 2.84 | 7.6 | 2.22 | 0.36 | 2.58 | 28.6 | 39.8 | 11.1 | 15.4 | — | 2.95 |
| 1934, June 13 | Rather small, green, very firm .. | .. | .. | 1.72 | 0.44 | 2.16 | — | — | — | — | — | — |
| 1936, " 8 | Large, green .. | .. | .. | 2.08 | 0.45 | 2.53 | 25.8 | — | 10.2 | — | — | — |
| " 17 | Mixed sizes .. | .. | .. | 1.54 | 0.26 | 1.80 | 16.2 | — | 9.0 | — | — | — |
| " Highest | .. | 3.49 | 10.2 | 2.49 | 0.45 | 2.92 | 29.9 | 40.9 | 11.7 | 15.4 | 1.50* | 3.40 |
| " Lowest | .. | 2.24 | 6.2* | 1.27* | 0.26 | 1.65 | 16.2 | 39.8 | 9.0 | — | 0.59 | 2.90 |
| " Average | .. | 2.66 | 7.81 | 2.00 | 0.37 | 2.37 | 25.0 | 40.4 | 10.36 | 15.4 | 0.84 | 3.08 |
| " No. of samples | .. | 5 | 15 | 21 | 21 | 21 | 19 | 2 | 19 | 2 | 11 | 18 |

TABLE IV—STRAWBERRIES

| Date | Description | Insol. solids Per Cent. | Seeds Per Cent. | Sol. solids Per Cent. | Acid as citric (cryst.) | | | Lead numbers | | Lead number per 1% total acid | | Pectin Per Cent. | pH (60% extract) |
|------------------|---|----------------------------|--------------------|--------------------------|-------------------------|-----------------------|--------------------|--------------|---------|----------------------------------|---------|---------------------|------------------------|
| | | | | | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Aqueous | Acetone | Aqueous | Acetone | | |
| 1930, June 17 | Large, fully ripe | — | — | 8.2 | 0.99 | 0.41 | 1.40 | 17.9 | — | 12.8 | — | 0.51 | 3.51 |
| " 18 | Partly ripe | — | — | 7.7 | 1.04 | 0.38 | 1.42 | 18.1 | — | 12.7 | — | 0.53 | 3.43 |
| " 20 | Mostly fully ripe, but firm | — | — | 9.3 | 1.08 | 0.35 | 1.43 | 17.9 | — | 12.5 | — | 0.68 | 3.36 |
| " 23 | Ripe, not fresh, slight mould | — | — | 7.6 | 1.05 | 0.33 | 1.38 | 18.3 | — | 13.3 | — | 0.43 | 3.36 |
| July 1 | Full ripe, ex Kent | — | — | 9.2 | 0.75 | 0.28 | 1.03 | 13.0 | — | 12.6 | — | 0.52 | 3.39 |
| " 2 | Large, full ripe | — | — | — | 0.95 | 0.37 | 1.32 | 15.5 | — | 11.7 | — | 0.50 | 3.49 |
| " 2 | Small, full ripe, not fresh | — | — | — | 0.74 | 0.37 | 1.11 | 12.5 | — | 11.3 | — | 0.39 | 3.71 |
| " 16 | Poor specimens, just ripe (end of season pickings) | — | — | — | 0.48 | 0.29 | 0.77 | 9.8 | — | 12.7 | — | — | 3.83 |
| " 21 | Medium size, just ripe | — | — | — | 0.93 | 0.38 | 1.31 | 16.7 | — | 12.7 | — | — | 3.49 |
| 1931, June 22 | Good size, not fully ripe, ex Kent | 2.05 | 0.91 | 9.3 | 1.24 | 0.35 | 1.59 | 16.7 | — | 10.5* | — | — | 3.17 |
| " 30 | Rather small, ripe and dry, but some mould | — | — | — | — | — | — | — | — | — | — | — | — |
| " 3 | Medium to small, some mould | 2.09 | 1.02 | 8.0 | 0.95 | 0.36 | 1.31 | 17.0 | — | 13.0 | — | — | 3.39 |
| 1932, June 28 | Rather small, fairly ripe, some crushed | 2.14 | 1.22 | 8.0 | 1.05 | 0.38 | 1.43 | 17.5 | — | 12.2 | — | — | 3.33 |
| " 28 | Mixed sizes, good condition | 2.63 | 1.40 | 9.0 | 1.30 | 0.42 | 1.76 | 22.7 | 27.0 | 15.3 | — | — | 3.38 |
| July 8 | Moderate size, mainly soft and ripe | 1.89 | 1.15 | 8.2 | 1.02 | 0.37 | 1.39 | 18.4 | 21.5 | 13.2 | 15.5 | — | 3.62 |
| " 5 | Medium size, mostly ripe | 1.84 | 0.76 | 7.6 | 0.71 | 0.30 | 1.01 | 13.0 | 15.8 | 12.9 | 15.6 | — | 3.63 |
| 1933, June 15 | Small, uniform, ripe | 2.27 | 1.27 | — | — | — | — | — | — | — | — | — | — |
| " 27 | Mixed sizes, mainly ripe, fresh | 1.96 | — | 10.1 | — | — | — | — | — | — | — | — | — |
| " 30 | Medium to small, dry | 1.92 | 1.11 | 9.4 | 0.89 | 0.41 | 1.30 | 16.1 | 19.0 | 12.4 | 14.6 | — | — |
| 1936 " 30 | Medium to small, dry | 2.02 | 1.19 | — | 1.00 | 0.42 | 1.42 | 18.0 | 21.3 | 12.7 | 15.0 | — | 3.45 |
| " 7 | Medium to small, some under-ripe | 2.32 | — | — | 0.78 | 0.32 | 1.10 | 14.4 | — | 13.1 | — | — | 3.54 |
| " 20 | Dutch Jucunda, rather under-ripe | 1.57 | — | 6.0 | 1.17 | 0.39 | 1.56 | 19.7 | 24.3 | 12.6 | 15.6 | — | 3.28 |
| 1937, June 21 | Dutch Evers, rather over-ripe, crushed and mouldy | 2.60 | — | 10.0 | 0.70 | 0.21 | 0.91 | 12.4 | 14.3 | 13.5 | 15.5 | 0.43 | 3.28 |
| " 25 | Dutch Jucunda, ripe, slight mould, rather sandy | 2.45 | — | 9.7 | 1.00 | 0.34 | 1.34 | 17.2 | 20.6 | 12.8 | 15.2 | 0.47 | 3.46 |
| " 30 | Dutch Jucunda, medium size, ripe, rather wet and not fresh | 2.11 | — | 8.8 | 0.93 | 0.38 | 1.31 | 18.2 | 20.6 | 13.8 | 15.6 | 0.63 | 3.48 |
| " 24 | Dutch Evers, small, ripe, some crushed, fairly fresh | 1.94 | — | 9.3 | 0.98 | 0.45 | 1.43 | 19.6 | 22.3 | 13.8 | 15.6 | 0.68 | 3.50 |
| July 2 | Dutch, small, ripe, bruised, slight mould, plugs taken out | 2.01 | — | 8.1 | 0.90 | 0.38 | 1.28 | 16.2 | 19.2 | 12.7 | 15.0 | 0.52 | 3.55 |
| " 22 | Dutch, small, very much crushed, many mouldy, plugs taken out | 2.59 | — | 7.8 | 0.88 | 0.38 | 1.26 | 16.8 | 19.7 | 13.2 | 15.5 | 0.53 | 3.48 |
| " Highest | .. | 2.63 | 1.40 | 10.1 | 1.30 | 0.45 | 1.72 | 22.7 | 27.0 | 13.8 | 15.6 | 0.68 | 3.83 |
| " Lowest | .. | 1.57 | 0.76 | 6.0 | 0.48 | 0.21 | 0.77 | 9.8 | 14.3 | 11.3 | 14.6 | 0.39 | 3.17 |
| " Average | .. | 2.11 | 1.11 | 8.56 | 0.95 | 0.36 | 1.31 | 18.67 | 20.68 | 12.77 | 15.31 | 0.53 | 3.46 |
| " No. of samples | .. | 19 | 9 | 20 | 26 | 26 | 26 | 25 | 13 | 25 | 13 | 14 | 25 |

* These results are doubtful and are not included in extremes and average.

TABLE V—RASPBERRIES

| Date | Description | Insol. solids | | | Seeds | | | Sol. solids | | | Acid as citric (cryst.) | | | Lead numbers | | | Lead number per 1% total acid | | | pH (50% extract) |
|--------------------|--|---------------|-----------|-----------|-----------|-----------|-----------|-------------|-----------|-----------|-------------------------|--------------------|-----------------|--------------|---------|-------|-------------------------------|---------|---|------------------|
| | | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Aqueous | Acetone | — | Aqueous | Acetone | — | |
| 1930, July 10 | Large, ripe, <i>ex</i> Kent | .. | .. | .. | .. | .. | .. | .. | .. | .. | 1.45 | 0.38 | 1.83 | 24.5 | — | — | 13.4 | — | — | 3.27 |
| " " 10 | Moderate size, ripe, <i>ex</i> Hants | .. | .. | .. | .. | .. | .. | .. | .. | .. | 1.96 | 0.37 | 2.33 | 33.1 | — | — | 14.2 | — | — | 3.04 |
| " " 15 | Fairly large, ripe, <i>ex</i> Kent | .. | .. | .. | .. | .. | .. | .. | .. | .. | 1.37 | 0.37 | 1.74 | 23.7 | — | — | 13.6 | — | — | 3.26 |
| " " 15 | Fairly large, ripe, <i>ex</i> Kent | .. | .. | .. | .. | .. | .. | .. | .. | .. | 1.87 | 0.40 | 1.97 | 25.9 | — | — | 13.1 | — | — | 3.28 |
| " " 22 | Medium size, ripe, fairly fresh | .. | .. | .. | .. | .. | .. | .. | .. | .. | 1.39 | 0.38 | 1.77 | 21.4 | — | — | 12.1 | — | — | 3.28 |
| " " 23 | Medium size, some under-ripe | .. | .. | .. | .. | .. | .. | .. | .. | .. | 1.24 | 0.31 | 1.55 | 19.7 | — | — | 12.7 | — | — | 3.20 |
| 1931, " 17 | Mixed sizes, mostly soft and rather mouldy, poor quality | .. | .. | .. | .. | .. | .. | .. | .. | .. | 1.02 | 0.42 | 1.44 | 19.0 | — | — | 13.2 | — | — | 3.46 |
| " " 29 | Large, some bruised, generally good | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.02 | 0.34 | 2.36 | 29.9 | — | — | 12.7 | — | — | 2.87 |
| 1932, " 14 | Large, crushed in lower layers, rather over-ripe | .. | .. | .. | .. | .. | .. | .. | .. | .. | 8.2 | 0.34 | 2.36 | 29.9 | — | — | 12.7 | — | — | 3.46 |
| " " 16 | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 7.8 | 1.04 | 0.45 | 34.2 | 37.4 | 14.3 | 15.6 | 3.22 | — | — |
| " " 20 | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 7.5 | 2.01 | 0.47 | 33.8 | 37.3 | 13.6 | 15.0 | 3.17 | — | — |
| 1933, " 25 | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 6.0 | 1.80 | 0.48 | 28.9 | 35.0 | 12.7 | 15.4 | 3.10 | — | — |
| 1935, Aug. 9 | Scottish | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.04 | — | — | — | — | — | — | — | — | — |
| 1936, July 28 | Medium size, rather over-ripe | .. | .. | .. | .. | .. | .. | .. | .. | .. | 1.69 | 0.43 | 2.12 | 28.9 | 32.2 | 13.6 | 15.2 | — | — | — |
| " " Highest | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.14 | 0.45 | 2.59 | 35.7 | 40.2 | 13.8 | 15.5 | 3.20 | — | — |
| " " Lowest | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 12.2* | 0.48 | 2.59 | 35.7 | 40.2 | 14.3 | 15.6 | 3.46 | — | — |
| " " Average | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 6.0 | 1.02* | 0.31 | 1.44 | 19.0 | 12.1 | 15.0 | 3.87 | — | — |
| " " No. of samples | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 8.37 | 1.71 | 0.41 | 20.7 | 27.7 | 13.36 | 15.34 | 3.90 | — | — |
| | | | | | | | | | | | 36 | 13 | 13 | 13 | 5 | 13 | 5 | 12 | — | — |

Embracing also analyses of 1934 season, already published (T. Macara*).

TABLE VI—REDCURRANTS

| Date | Description | Insol. solids | | | Seeds | | | Sol. solids | | | Acid as citric (cryst.) | | | Lead numbers | | | Lead number per 1% total acid | | | Pectin Per Cent. | pH (50% extract) |
|----------------|--|---------------|-----------|-----------|-----------|-----------|-----------|-------------|-----------|-----------|-------------------------|--------------------|-----------------|--------------|---------|-------|-------------------------------|---------|---|------------------|------------------|
| | | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Per Cent. | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Aqueous | Acetone | — | Aqueous | Acetone | — | | |
| 1930, July 16 | Medium to large, ripe, good condition | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.12 | 0.35 | 2.47 | 35.4 | — | — | 14.3 | — | — | — | 3.05 |
| " " 20 | Medium to large, ripe, rather moist but otherwise good | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.70 | 0.39 | 3.09 | 43.1 | — | — | 14.0 | — | — | — | 3.02 |
| " " 23 | Slightly dry | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.54 | 0.42 | 2.96 | 35.4 | — | — | 12.0* | — | — | — | 3.03 |
| " " 29 | Large, fully ripe, wet with juice | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.40 | 0.45 | 2.85 | 37.7 | — | — | 13.2 | — | — | — | 3.07 |
| Aug. 5 | Ripe, but not fresh, wet condition | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.40 | 0.42 | 2.82 | 34.9 | — | — | 12.4 | — | — | — | 2.95 |
| 1931, June 26 | Fairly large, ripe, rather crushed | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.06 | 0.41 | 2.47 | 34.8 | — | — | 14.1 | — | — | — | 3.10 |
| 1932, " 7 | Moderate size, rather under-ripe, dry | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.48 | 0.49 | 2.97 | 41.7 | 50.7 | 17.1 | 0.73 | — | — | — | 3.02 |
| " " 15 | Kentish | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.67 | 0.43 | 3.10 | — | 51.8 | 16.6 | — | — | — | — | 3.05 |
| 1934, " 17 | Large, ripe, good condition | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.73 | — | — | — | — | — | — | — | — | — | 3.15 |
| " " 18 | Large, fresh condition, very few damaged | .. | .. | .. | .. | .. | .. | .. | .. | .. | — | — | — | — | — | — | — | — | — | — | — |
| 1938, " 29 | Norfolk, medium size, ripe, good condition | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.60 | 0.50 | 3.10 | 41.8 | 49.7 | 13.5 | 16.0 | — | — | — | — |
| " " Aug. 9 | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | — | — | — | — | — | — | 13.5 | 15.3 | — | — | — |
| Highest | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.73 | 0.50 | 3.10 | 43.1 | 51.8 | 14.3 | 17.1 | 0.73* | — | — | 3.22 |
| Lowest | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 2.06* | 0.35 | 2.47 | 34.8 | 49.7 | 12.4 | 15.3 | 0.47 | — | — | 2.95 |
| Average | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 6.02 | 4.50 | 10.23 | 2.46 | 50.7 | 13.64 | 16.25 | 0.60 | — | — | 3.07 |
| No. of samples | .. | .. | .. | .. | .. | .. | .. | .. | .. | .. | 3 | 6 | 9 | 3 | 8 | 4 | 2 | 10 | — | — | — |

* These results are doubtful and are not included in extremes and average.

TABLE VII—BLACKCURRANTS

| Date | Description | Insol. solids Per Cent. | Seeds Per Cent. | Sol. solids Per Cent. | Acid as citric (cryst.) | | | Lead numbers | | Lead number per 1% total acid | | Pectin Per Cent. | pH (60% extract) |
|---------------|--|----------------------------|--------------------|--------------------------|-------------------------|-----------------------|--------------------|--------------|---------|-------------------------------|---------|---------------------|---------------------|
| | | | | | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Aqueous | Acetone | Aqueous | Acetone | | |
| 1930, July 17 | Medium size, ripe .. | — | — | — | 3.66 | 0.67 | 4.33 | 58.9 | — | 13.6 | — | — | 3.10 |
| " " 21 | Mixed sizes, rather dirty .. | — | 2.89 | — | 2.67 | 0.58 | 3.25 | 39.5 | — | 12.2 | — | — | 3.18 |
| " " 24 | Fairly large, ripe, not fresh .. | — | 4.35 | — | 3.38 | 0.52 | 3.90 | 51.3 | — | 13.1 | — | — | 3.02 |
| " Aug. 2 | Mixed sizes, slightly moist with juice .. | — | 3.94 | — | 2.73 | 0.45 | 3.18 | 42.6 | — | 13.4 | — | — | 3.06 |
| 1931, June 16 | Belgian, very mixed size, poor condition .. | 6.34 | 3.01 | 10.6 | 3.11 | 0.70 | 3.81 | 50.7 | — | 13.3 | — | — | 3.20 |
| " " 18 | Very small to medium size, fairly ripe, not fresh .. | 5.98 | 2.33 | 10.9 | 3.18 | 0.67 | 3.85 | 50.8 | — | 13.2 | — | — | 2.98 |
| " " 25 | Medium to small, fairly ripe and fresh .. | 7.87 | 4.00 | 11.3 | 2.85 | 0.67 | 3.52 | 46.0 | — | 13.1 | — | — | 3.22 |
| " July 14 | Medium size, very soft, some crushed .. | 6.79 | 3.12 | 12.8 | 3.57 | 0.72 | 4.29 | 57.8 | — | 13.5 | — | — | 3.07 |
| 1932, " 13 | Size varied, fair proportion under-ripe .. | 5.98 | — | 8.9 | 3.69 | 0.80 | 4.49 | 64.5 | 74.1 | 14.2 | 16.3 | — | 3.17 |
| " " 20 | — .. | 5.77 | 3.54 | 9.2 | 3.80 | 0.74 | 4.54 | 64.8 | 78.2 | 14.4 | 17.3 | — | 2.94 |
| 1933, " 5 | — .. | 5.53 | — | 13.6 | 3.40 | — | — | — | — | — | — | — | — |
| " " 20 | — .. | 5.33 | — | 13.6 | 3.85 | — | — | — | — | — | — | — | — |
| 1934, Aug. 2 | Large, fresh condition, dry .. | — | — | — | 3.40 | 0.61 | 4.01 | — | — | — | — | 0.94 | 3.17 |
| 1936, July 27 | Large, ripe .. | 4.11 | — | — | 2.93 | 0.65 | 3.58 | 46.5 | 57.3 | 13.0 | 16.0 | — | 3.18 |
| " " 28 | Large, ripe .. | 4.74 | — | — | 2.99 | 0.53 | 3.52 | 46.1 | 54.6 | 13.1 | 15.5 | — | 2.94 |
| " Aug. 8 | Medium size, over-ripe .. | 4.94 | — | — | 2.66 | 0.52 | 3.18 | 39.7 | 49.9 | 12.5 | 15.7 | — | 3.08 |
| " " 11 | — .. | 5.44 | — | — | 3.71 | 0.72 | 4.43 | 58.3 | 67.7 | 13.2 | 15.3 | 1.19 | 3.10 |
| 1937, July 16 | Dutch, many very large, firm but ripe .. | 5.24 | — | 13.6 | 3.03 | 0.66 | 3.69 | 48.7 | 60.5 | 13.2 | 16.4 | 1.47 | 3.18 |
| " " 23 | Dutch, very large, not over-ripe, but rather soft and moist with juice .. | 4.26 | — | 11.5 | 2.42 | 0.58 | 3.00 | 40.9 | 48.1 | 13.7 | 16.0 | 1.04 | 3.09 |
| 1938, June 21 | French, rather small, some unripe, not very fresh, heavily sprayed (lime-sulphur) .. | 5.13 | — | 13.3 | 3.98 | 0.71 | 4.69 | 63.3 | 75.3 | 13.5 | 16.1 | 1.61 | 3.18 |
| " " 23 | French, moderate size, fairly ripe, some mouldy and crushed .. | 5.68 | — | 11.3 | 3.86 | 0.69 | 4.55 | 60.7 | 73.0 | 13.4 | 16.0 | 1.66 | 3.14 |
| " July 22 | Dutch, medium size, ripe, good condition .. | 5.46 | — | 15.4 | 2.67 | 0.66 | 3.33 | 48.9 | 55.7 | 14.7 | 16.8 | 1.41 | 3.29 |
| " — | Latvian, canned .. | — | — | — | — | — | — | — | — | 13.9 | 16.5 | — | — |
| " — | Estonian, canned .. | — | — | — | — | — | — | — | — | 13.8 | 16.5 | — | — |
| " — | Canadian, canned .. | — | — | — | — | — | — | — | — | 14.4 | 17.1 | — | — |
| " — | Highest .. | 7.87 | 4.35 | 15.4 | 3.98 | 0.80 | 4.69 | 64.8 | 78.2 | 14.7 | 17.3 | 1.66 | 3.29 |
| " — | Lowest .. | 4.11* | 2.33 | 8.9* | 2.42* | 0.45 | 3.00 | 39.5 | 48.1 | 12.2 | 15.3 | 0.94 | 2.94 |
| " — | Average .. | 5.57 | 3.40 | 11.99 | 3.25 | 0.64 | 3.86 | 51.6 | 63.1 | 13.47 | 16.25 | 1.33 | 3.11 |
| " — | No. of samples .. | 17 | 8 | 13 | 22 | 20 | 20 | 19 | 11 | 22 | 14 | 7 | 20 |

TABLE VIII—PLUMS

| Date | Description | Insol. solids | | | | Sol. solids | | | | Acid as citric (cryst.) | | | | Lead numbers | | Lead number per 1% total acid | | pH (50% extract) |
|--------------------|--|---------------|--------------|-------------|-----------|----------------|--------------------|-----------------|-----------|-------------------------|-------------------|-------------------|-------------------|--------------|---------|-------------------------------|---------|------------------|
| | | Insol. Cent. | Stones Cent. | † Per Cent. | Per Cent. | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Per Cent. | Aqueous Per Cent. | Acetone Per Cent. | Aqueous Per Cent. | Acetone Per Cent. | Aqueous | Acetone | Aqueous | Acetone | |
| 1930, July 28 | Italian, blue, medium size, moderately ripe | 1-16 | 4-5 | — | — | 2-11 | 0-40 | 2-51 | — | — | — | — | — | — | — | — | — | 2-94 |
| " Aug. 6 | Fully ripe, some slightly crushed | 1-19 | 4-5 | — | — | 1-82 | 0-37 | 2-19 | — | — | — | — | — | — | — | — | — | 2-93 |
| " " 18 | Blue-green, small, ripe | 1-57 | 4-3 | — | — | 1-72 | 0-52 | 2-24 | — | — | — | — | — | — | — | — | — | 3-06 |
| " " 25 | Victoria, mixed sizes, unequally ripe, rather crushed | 2-62 | 4-3 | — | — | 1-21 | 0-36 | 1-57 | — | — | — | — | — | — | — | — | — | 3-14 |
| " " 29 | Medium size, fully ripe | 1-32 | 4-2 | — | — | 1-32 | 0-30 | 1-62 | — | — | — | — | — | — | — | — | — | 3-01 |
| " " Sept. 1 | Red, large, ripe, but condition poor | 0-98 | 2-9 | — | — | 1-32 | 0-29 | 1-61 | — | — | — | — | — | — | — | — | — | 2-94 |
| " " 4 | Golden, large, ripe to over-ripe, some slightly bruised | 1-23 | 4-0 | — | — | 1-78 | 0-32 | 2-10 | — | — | — | — | — | — | — | — | — | 2-90 |
| " " 10 | Red, over-ripe, bruised and wet with juice | 0-85 | 4-0 | — | — | 1-53 | 0-35 | 1-88 | — | — | — | — | — | — | — | — | — | 3-02 |
| 1931, Aug. 5 | Blue, medium size, fully ripe | 1-29 | 5-4 | — | — | 1-16 | 0-34 | 1-50 | — | — | — | — | — | — | — | — | — | 3-13 |
| " " 31 | Victoria, fairly large, mixed ripe, over-ripe and green | 1-36 | 5-5 | 12-1 | — | 1-94 | 0-47 | 2-41 | — | — | — | — | — | — | — | — | — | 2-91 |
| " " Sept. 2 | Victoria, mixed sizes, ripe, bruised | 1-31 | 2-8 | 11-2 | — | 1-61 | 0-35 | 1-96 | — | — | — | — | — | — | — | — | — | 2-92 |
| " " 4 | Golden, very uneven in size and ripeness, fair condition | 1-31 | 4-6 | 10-4 | — | 1-65 | 0-36 | 2-00 | — | — | — | — | — | — | — | — | — | 2-88 |
| " " 14 | Red, small to medium size, ripe | 1-25 | 3-9 | 12-1 | — | 1-68 | 0-37 | 2-05 | 6-9* | — | — | — | — | — | — | 3-4* | — | 2-97 |
| 1932, " 1 | Blue type, hard and greenish | 1-76 | 3-8 | 16-6 | — | 2-62 | 0-43 | 3-05 | 7-6* | — | — | — | — | — | — | 2-5* | — | 2-80 |
| " " 14 | Victoria, Kentish, fairly large, ripe and sound | 1-15 | 3-7 | 11-7 | — | 1-31 | 0-23 | 1-54 | 9-7 | 24-9 | — | — | — | — | — | 5-5 | 14-2 | 3-08 |
| " " 22 | Blue type, mainly ripe | 1-39 | 4-0 | 15-6 | — | 1-84 | 0-51 | 2-35 | 8-6 | 19-0 | — | — | — | — | — | 5-6 | 12-3 | 2-82 |
| " " Highest | | 2-62* | 5-5 | 16-6 | — | 2-62 | 0-52 | 3-05 | 18-0 | 34-4 | — | — | — | — | — | 7-6 | 14-6 | 3-15 |
| " " Lowest | | 0-86 | 2-8 | 10-4 | — | 1-16 | 0-23 | 1-50 | 8-6 | 19-0 | — | — | — | — | — | 5-5* | 12-3 | 2-80 |
| " " Average | | 1-38 | 4-11 | 12-72 | — | 1-65 | 0-37 | 2-02 | 12-1 | 26-1 | — | — | — | — | — | 6-2 | 13-7 | 2-97 |
| " " No. of samples | | 17 | 17 | 8 | — | 17 | 17 | 17 | 3 | 3 | — | — | — | — | — | 3 | 3 | 17 |

* These results are doubtful and are not included in extraneous and average.

TABLE IX—GREENGAGES

| Date | Description | Insol. solids | | | | Sol. solids | | | | Acid as citric (cryst.) | | | | Lead numbers | | Lead number per 1% total acid | | pH (50% extract) |
|--------------------|--|---------------|--------------|-------------|-----------|----------------|--------------------|-----------------|-----------|-------------------------|-------------------|-------------------|-------------------|--------------|---------|-------------------------------|---------|------------------|
| | | Insol. Cent. | Stones Cent. | † Per Cent. | Per Cent. | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Per Cent. | Aqueous Per Cent. | Acetone Per Cent. | Aqueous Per Cent. | Acetone Per Cent. | Aqueous | Acetone | Aqueous | Acetone | |
| 1930, July 25 | Small, hard, unripe | 2-93 | 5-0 | — | — | 1-49 | 0-54 | 2-03 | — | — | — | — | — | — | — | — | — | 3-08 |
| " Aug. 7 | Rather small, ripe | 1-96 | 5-2 | — | — | 1-12 | 0-50 | 1-62 | — | — | — | — | — | — | — | — | — | 3-13 |
| " " 13 | Mixed in size, ripe, bruised | 2-33 | 5-1 | — | — | 1-25 | 0-44 | 1-69 | — | — | — | — | — | — | — | — | — | 3-12 |
| " Sept. 4 | English, small, firm, ripe, not fresh | 1-41 | 7-1 | — | — | 1-17 | 0-41 | 1-58 | — | — | — | — | — | — | — | — | — | 3-16 |
| 1931, July 27 | Italian, mostly sound and ripe | 1-14 | 6-5 | 9-8 | — | 1-41 | 0-41 | 1-82 | — | — | — | — | — | — | — | — | — | 3-06 |
| " Aug. 7 | Medium size, full ripe | 1-98 | 5-3 | 17-2 | — | 1-16 | 0-64 | 1-80 | — | — | — | — | — | — | — | — | — | 3-21 |
| 1932, Sept. 5 | English, ripe, soft, a little bruised and some mould | 1-38 | 2-9 | 15-5 | — | 1-73 | 0-44 | 2-17 | 11-9 | 28-2 | — | — | — | — | — | 5-5 | 13-0 | 3-00 |
| " " 12 | English, large, ripe and soft | 1-14 | 3-5 | 13-2 | — | 1-01 | 0-36 | 1-37 | 8-1 | 17-1 | — | — | — | — | — | 5-9 | 12-5 | 3-19 |
| " " 21 | — | — | — | — | — | 0-96 | 0-49 | 1-45 | — | — | — | — | — | — | — | — | — | 3-46 |
| 1936, " 18 | English, medium size, rather unripe | — | — | — | — | 1-97 | 0-43 | 2-40 | 16-6 | 34-1 | — | — | — | — | — | 6-9 | 14-2 | — |
| " " 25 | — | — | — | — | — | 2-20 | 0-56 | 2-76 | 17-1 | 38-1 | — | — | — | — | — | 6-2 | 13-8 | — |
| " " Highest | | 2-93* | 7-1 | 17-2 | — | 2-20* | 0-64 | 2-76 | 17-1 | 38-1 | — | — | — | — | — | 6-9 | 14-2 | 3-46 |
| " " Lowest | | 1-14 | 2-9* | 9-8* | — | 0-96* | 0-36 | 1-37 | 8-1 | 15-0 | — | — | — | — | — | 5-5 | 10-3 | 3-00 |
| " " Average | | 1-79 | 5-1 | 13-91 | — | 1-41 | 0-43 | 1-89 | 13-4 | 26-6 | — | — | — | — | — | 6-12 | 12-74 | 3-16 |
| " " No. of samples | | 8 | 8 | 4 | — | 11 | 11 | 11 | 4 | 5 | — | — | — | — | — | 4 | 5 | 9 |

† On entire fruit; other data on stone-free portion only.

TABLE X—DAMSONS

| Date | Description | Insol. solids Per Cent. | Stones Per Cent. | Sol. solids Per Cent. | Acid as citric (cryst.) | | | Lead numbers | | Lead number per 1% total acid | | pH (50% extract) |
|---------------|--|-------------------------|------------------|-----------------------|-------------------------|--------------------|-----------------|--------------|---------|-------------------------------|---------|------------------|
| | | | | | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Aqueous | Acetone | Aqueous | Acetone | |
| 1930, Sept. 1 | Rather small, ripe and sound | 1.92 | 8.4 | — | 2.53 | 0.58 | 3.11 | — | — | — | — | 3.01 |
| " 5 | Small, ripe | 1.74 | 8.9 | — | 2.39 | 0.65 | 3.04 | — | — | — | — | 3.01 |
| " 8 | Mostly small, ripe, not fresh | 2.26 | 9.9 | — | 2.21 | 0.63 | 2.84 | — | — | — | — | 3.11 |
| " 12 | Medium size, ripe, not quite fresh | 2.15 | 10.8 | — | 2.05 | 0.60 | 2.65 | — | — | — | — | 3.10 |
| " 23 | Large, fully ripe or over-ripe, and wet with juice | 2.18 | 7.3 | — | 2.01 | 0.63 | 2.64 | — | — | — | — | 3.12 |
| 1931, " 6 | Medium to small, firm and fresh, sour flavour | 2.50 | 6.8 | 13.6 | 2.26 | 0.53 | 2.79 | 9.8* | — | 3.5* | — | 2.96 |
| " 16 | Mostly small, ripe, not fresh | 2.39 | 7.7 | 13.0 | 2.49 | 0.64 | 3.13 | 9.4* | — | 3.0* | — | 3.00 |
| 1932, " 20 | Rather over-ripe | 2.43 | 5.8 | 15.6 | 2.14 | 0.64 | 2.78 | 19.4 | 43.3 | 7.0 | 15.6 | 3.19 |
| " Oct. 3 | Californian, rather large, some soft and over-ripe | 1.44 | 4.6 | 14.8 | 2.01 | 0.38 | 2.39 | 13.3 | 34.9 | 5.6 | 14.6 | 2.98 |
| | Highest | 2.50 | 10.8 | 15.6 | 2.53 | 0.65 | 3.13 | 19.4 | 43.3 | 7.0 | 15.6 | 3.19 |
| | Lowest | 1.44 | 4.6 | 13.0 | 2.01 | 0.38 | 2.39 | 13.3 | 34.9 | 5.6 | 14.6 | 2.98 |
| | Average | 2.11 | 7.8 | 14.25 | 2.23 | 0.59 | 2.82 | 16.4 | 39.1 | 6.3 | 15.1 | 3.05 |
| | No. of samples | 9 | 9 | 4 | 9 | 9 | 9 | 2 | 2 | 2 | 2 | 9 |

† On entire fruit; other data on stone-free portion only.

* These results are doubtful and are not included in extremes and average.

TABLE XI—APPLES

| Date | Description | Insol. solids Per Cent. | Sol. solids Per Cent. | Acid as citric (cryst.) | | | Lead numbers | | Lead numbers per 1% total acid | | pH (50% extract) |
|------------------|--|-------------------------|-----------------------|-------------------------|--------------------|-----------------|--------------|---------|--------------------------------|---------|------------------|
| | | | | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Aqueous | Acetone | Aqueous | Acetone | |
| 1930, July 25 | Hard, green | .. | .. | 1.61 | 0.24 | 1.85 | — | — | — | — | 2.86 |
| " 30 | Hard, green, sour | .. | .. | 1.37 | 0.19 | 1.56 | — | — | — | — | 2.86 |
| " Aug. 6 | Small, green | .. | .. | 1.27 | 0.28 | 1.55 | — | — | — | — | 3.00 |
| " 7 | Large, green, sour | .. | .. | 1.61 | 0.22 | 1.83 | — | — | — | — | 2.80 |
| " 15 | Large, green, bruised | .. | .. | 1.34 | 0.24 | 1.58 | — | — | — | — | 2.92 |
| " 27 | Small, green, bruised | .. | .. | 1.24 | 0.20 | 1.44 | — | — | — | — | 2.89 |
| Sept. 3 | Medium size, beginning to ripen, bruised | .. | .. | 0.96 | 0.25 | 1.21 | — | — | — | — | 3.11 |
| " 8 | Medium size, dark green, hard, bruised, sweet taste | .. | .. | 0.82 | 0.22 | 1.04 | — | — | — | — | 3.16 |
| " 19 | Very large, beginning to reddish and ripen, slightly bruised | .. | .. | 0.94 | 0.22 | 1.16 | — | — | — | — | 3.10 |
| " 29 | Medium size, fairly ripe, slightly bruised | .. | .. | 0.82 | 0.30 | 1.12 | — | — | — | — | 3.38 |
| " Oct. 30 | Medium size, green, but fairly ripe | .. | .. | 0.61 | 0.28 | 0.89 | — | — | — | — | 3.45 |
| 1932, Aug. 30 | Kentish | 2.21 | 9.8 | 1.21 | 0.24 | 1.45 | 10.9 | 22.2 | 7.4 | 15.1 | 2.96 |
| " Sept. 9 | Hard, yellow-green | 2.09 | 11.2 | 1.15 | 0.28 | 1.43 | 10.1 | 21.2 | 7.1 | 14.9 | 3.09 |
| " 16 | Bramleys, rather large, green, some rather sweet | 2.61 | 11.1 | 1.08 | 0.24 | 1.32 | 9.7 | 19.6 | 7.4 | 15.0 | 3.13 |
| " 29 | Rather large, green, fairly sweet | 3.20 | 11.7 | 0.82 | 0.28 | 1.10 | 6.9 | 16.5 | 6.3 | 15.0 | 3.22 |
| | Highest | 3.20 | 11.7 | 1.61 | 0.30 | 1.85 | 10.9 | 22.2 | 7.4 | 15.1 | 3.45 |
| " 9 | Lowest | 2.09 | 9.8 | 0.61 | 0.19 | 0.89 | 6.9 | 16.5 | 6.3 | 14.9 | 2.80 |
| " Average | .. | 2.53 | 11.0 | 1.12 | 0.25 | 1.37 | 9.4 | 19.9 | 7.05 | 15.0 | 3.06 |
| " No. of samples | .. | 4 | 4 | 15 | 15 | 15 | 4 | 4 | 4 | 4 | 15 |

TABLE XII—BILBERRIES

| Date | Description | Insol. solids | | Seeds | | Acid as citric (cryst.) | | | | Lead number per 1% (Aqueous) | Pectin Per Cent. | pH (50% extract) |
|----------------|--|---------------|-------|-----------|-------|-------------------------|--------------------|-----------------|-------|------------------------------|------------------|------------------|
| | | Per Cent. | Cent. | Per Cent. | Cent. | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Cent. | | | |
| 1929, Sept. 29 | Cumberland (a small loss of juice occurred in transit) | 3.87 | — | — | — | 1.10 | — | — | — | — | 0.37 | — |
| 1930, Aug. 26 | Norwegian, fully ripe, good condition | 3.96 | 2.31 | — | — | 1.21 | 0.24 | 1.45 | — | 11.6 | 0.51 | 2.95 |

TABLE XIII—BLACKBERRIES

| Date | Description | Insol. solids | | Seeds | | Acid as citric (cryst.) | | | | Lead number per 1% total acid | | Pectin Per Cent. | pH (50% extract) |
|----------------------|--|---------------|-------|-----------|-------|-------------------------|--------------------|-----------------|-------|-------------------------------|---|------------------|------------------|
| | | Per Cent. | Cent. | Per Cent. | Cent. | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Cent. | | | | |
| 1930, Aug. 25 | Large, firm and black, but not fully ripe | — | — | 4.34 | — | 2.26 | 0.34 | 2.60 | 22.5 | 8.7 | — | — | 2.64 |
| " Sept. 5 | Large, hedgerow fruit, uneven ripeness | 7.44 | — | 5.56 | — | 1.20 | 0.37 | 1.57 | 14.5 | 9.2 | — | — | 3.12 |
| " " 11 | Hedgerow (Middlesex), medium size, firm, uneven ripeness | 9.72 | — | 7.30 | — | 1.75 | 0.47 | 2.22 | 21.6 | 9.7 | — | — | 2.95 |
| " " 19 | Small, fairly ripe, wet with rain | 9.55 | — | 8.01 | — | 0.48 | 0.45 | 0.93 | 9.5 | 10.2 | — | — | 3.83 |
| " " 22 | Medium size, fully ripe | 9.15 | — | 7.65 | — | 0.72 | 0.45 | 1.17 | 11.9 | 10.2 | — | — | 3.60 |
| " " 23 | Small, not very ripe, rather wet | 9.47 | — | 7.63 | — | 0.79 | 0.52 | 1.31 | 11.6 | 8.9 | — | — | 3.62 |
| " " 30 | Large, uneven ripeness, wet with rain | 8.44 | — | — | — | 1.09 | 0.56 | 1.65 | 13.3 | 8.1 | — | — | 3.39 |
| " Oct. 7 | Medium size, full ripe, slightly wet with juice | 10.74 | — | 8.88 | — | 0.60 | 0.47 | 1.07 | 10.0 | 9.3 | — | — | 3.79 |
| 1931, Sept. 22 | Rather small, fairly ripe and fresh | 9.21 | 8.1 | 6.93 | 8.1 | 0.36 | 0.47 | 0.83 | 7.2 | 8.6 | — | — | 4.21 |
| " Oct. 1 | Medium size, barely ripe, some mould | 9.64 | 7.9 | 7.26 | 7.9 | 0.64 | 0.49 | 1.13 | 10.1 | 8.9 | — | — | 3.86 |
| 1932, Sept. 24 | Large, fair condition, but some mouldy | 5.05 | 7.7 | 3.79 | 7.7 | 1.82 | 0.40 | 2.22 | 19.4 | 34.4 | — | — | 3.16 |
| " " 28 | Rather large, slightly under-ripe, fresh condition | 7.86 | — | — | — | 1.03 | 0.43 | 1.46 | 14.6 | 22.2 | — | — | 3.29 |
| " " " | Highest | 10.74 | 8.88 | 8.88 | 8.2 | 2.26* | 0.56 | 2.60 | 22.5 | 34.4 | — | — | 4.21 |
| " " " | Lowest | 5.05* | 3.79 | 7.7* | 7.7* | 0.36* | 0.34 | 0.83 | 7.2 | 22.2 | — | — | 2.64 |
| " " " | Average | 8.75 | 6.73 | 8.0 | 8.0 | 1.06 | 0.45 | 1.51 | 13.9 | 28.3 | — | — | 3.46 |
| " " " | No. of samples | 11 | 10 | 4 | 4 | 12 | 12 | 12 | 12 | 2 | — | — | 12 |
| CANNED BLACKBERRIES. | | | | | | | | | | | | | |
| 1934 — | Solid Pack, U.S.A. | 8.56 | — | — | 10.35 | 0.69 | 0.31 | 1.00 | 10.0 | 10.0 | — | 0.56 | — |

TABLE XIV—APRICOTS

| Date | Description | Insol. solids Per Cent. | † Stones Per Cent. | Acid as citric (cryst.) | | | | Lead numbers | | Lead number per 1% total acid | | Pectin Per Cent. | pH (60% extract) |
|--------------|-----------------|----------------------------|--------------------------|--------------------------|-------------------|-----------------------|--------------------|--------------|---------|----------------------------------|---------|---------------------|------------------------|
| | | | | Sol. solids Per Cent. | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Aqueous | Acetone | Aqueous | Acetone | | |
| 1932, July 1 | — | 1.16 | 5.5 | 9.9 | 0.92 | 0.62 | 1.54 | 14.7 | 23.5 | 9.6 | 15.3 | — | 3.90 |
| 1938, " 2 | Italian, ripe | 1.58 | — | 14.0 | 2.02 | 0.67 | 2.69 | 27.0 | 40.1 | 10.0 | 15.2 | 0.88 | 3.46 |
| " " 2 | Italian, unripe | 1.57 | — | 12.4 | 2.15 | 0.63 | 2.78 | — | — | — | — | 1.04 | 3.42 |
| 1939, Jan. 6 | South African | 1.78 | 6.5 | 15.7 | 1.19 | 0.59 | 1.78 | — | — | — | — | 0.90 | 3.86 |
| | Highest | 1.78 | 6.5 | 15.7 | 2.15 | 0.67 | 2.78 | 27.0 | 40.1 | 10.0 | 15.3 | 1.04 | 3.90 |
| | Lowest | 1.16 | 5.5 | 9.9 | 0.92 | 0.59 | 1.54 | 14.7 | 23.5 | 9.6 | 15.2 | 0.88 | 3.42 |
| | Average | 1.52 | 6.0 | 13.0 | 1.57 | 0.63 | 2.20 | 20.8 | 31.8 | 9.8 | 15.25 | 0.94 | 3.66 |
| | No. of samples | 4 | 2 | 4 | 4 | 4 | 4 | 2 | 2 | 2 | 2 | 3 | 4 |

† On entire fruit; other data on stone-free portion only.

TABLE XV—APRICOT PULPS

| Date | Description | Insol. solids Per Cent. | † Stones Per Cent. | Sol. solids Per Cent. | Acid as citric (cryst.) | | | Lead numbers | | Lead number per 1% total acid | | Pectin Per Cent. | pH (60% extract) |
|---------------|---------------------------|----------------------------------|-----------------------------|--------------------------------|-------------------------|--------------------------|-----------------------|--------------|---------|----------------------------------|---------|------------------------|------------------------|
| | | | | | Free Per Cent. | Combined Per Cent. | Total Per Cent. | Lead numbers | | Aqueous | | | |
| | | | | | | | | Aqueous | Acetone | Aqueous | Acetone | | |
| 1932, Nov. 8 | — | 1.24 | — | 10.7 | 1.36 | 0.54 | 1.90 | 20.8 | 25.9 | 11.0 | 13.6 | 0.58 | 3.45 |
| 1937, Oct. 2 | Average pasty consistency | 1.00 | — | 14.6 | 1.80 | 0.38 | 2.18 | 24.0 | — | 11.0 | — | — | — |
| " " 2 | Average pasty consistency | 1.20 | — | 13.0 | 2.05 | 0.63 | 2.68 | 27.0 | — | 10.1 | — | — | — |
| " " 2 | Very thick and fruity | 1.66 | — | 11.8 | 2.64 | 0.55 | 3.19 | 33.0 | — | 10.4 | — | — | — |
| " " 2 | Average pasty consistency | 1.15 | — | 16.2 | 1.33 | 0.63 | 1.96 | 22.4 | — | 11.4 | — | — | — |
| 1938, Feb. 22 | South African | 1.41 | — | 16.4 | 1.70 | 0.56 | 2.26 | 20.3 | 32.8 | 9.0 | 14.7 | 0.81 | — |
| " Aug. 23 | Spanish (casks) | 0.92 | — | 12.0 | 2.17 | 0.30 | 2.47 | 28.2 | 37.5 | 11.4 | 15.2 | 0.59 | 2.86 |
| " " 23 | Spanish (casks) | 0.89 | — | 12.5 | 2.17 | 0.33 | 2.50 | 28.3 | 38.3 | 11.3 | 15.3 | 0.68 | 2.89 |
| " " 23 | Syrian (casks) | 1.73 | — | 13.0 | 1.83 | 0.68 | 2.51 | 27.2 | 36.1 | 10.8 | 14.4 | 0.69 | 3.41 |
| " " 23 | Syrian (casks) | 1.54 | — | 13.0 | 1.69 | 0.73 | 2.42 | 27.4 | 37.1 | 11.3 | 15.3 | 0.78 | 3.44 |
| | Highest | 1.73 | — | 16.4 | 2.64 | 0.73 | 3.19 | 33.0 | 38.3 | 11.4 | 15.3 | 0.81 | 3.45 |
| | Lowest | 0.89 | — | 10.7 | 1.33 | 0.30 | 1.90 | 20.3 | 25.9 | 9.0 | 13.6 | 0.58 | 2.86 |
| | Average | 1.27 | — | 13.32 | 1.87 | 0.53 | 2.40 | 25.86 | 34.62 | 10.77 | 14.75 | 0.69 | 3.20 |
| | No. of samples | 10 | — | 10 | 10 | 10 | 10 | 10 | 6 | 10 | 6 | 6 | 5 |

Not determined when present; other data on stone-free portion only.

TABLE XVI—EFFECT OF REMOVAL OF COLOUR ON THE LEAD NUMBERS OF REDCURRANT EXTRACTS

| No. | | | | | | | Lead No. per 1 per cent. of total acid | |
|-----|------------------------|----|----|----|----|----|--|---------|
| | | | | | | | Aqueous | Acetone |
| 1 | Before decolorising .. | .. | .. | .. | .. | .. | 13.5 | 16.0 |
| | After .. | .. | .. | .. | .. | .. | 13.2 | 15.2 |
| 2 | Before .. | .. | .. | .. | .. | .. | 13.5 | 15.3 |
| | After .. | .. | .. | .. | .. | .. | 13.3 | 14.8 |

The aqueous lead numbers (per 1 per cent. of acid) indicate that the average proportion of malic acid in this fruit is practically nil. In one sample it appeared to be as high as 20 per cent., but this again was one of the early analyses by the lead method, and is open to some doubt.

Blackcurrants.—The minimum figures for insoluble and soluble solids and free acid are lower than before¹ and the averages for soluble solids and free acid are also appreciably lower.

The proportion of seeds in the insoluble matter varies from 39 per cent. to 61 per cent., the average being 49 per cent.

As with red currants, the lead numbers per 1 per cent. of acid exceed the figures to be expected from citric acid. This again appears to be due to lead-precipitating effects of the colouring matter. The lead numbers and acidities were determined on the extracts of the last three samples after treatment with decolorising carbon. Appreciably lower figures for the lead numbers per 1 per cent. of acid were found, and the acetone lead numbers were now much nearer the figure for citric acid or citric and malic acids. (Table XVII)

TABLE XVII—EFFECT OF REMOVAL OF COLOUR ON THE LEAD NUMBERS OF BLACKCURRANT EXTRACTS

| No. | | | | | | | Lead No. per 1 per cent. of total acid | |
|-----|---------------------|----|----|----|----|----|--|---------|
| | | | | | | | Aqueous | Acetone |
| 1 | Before decolorising | .. | .. | .. | .. | .. | 13.9 | 16.5 |
| | After .. | .. | .. | .. | .. | .. | 12.4 | 15.0 |
| 2 | Before .. | .. | .. | .. | .. | .. | 13.8 | 16.5 |
| | After .. | .. | .. | .. | .. | .. | 13.0 | 15.3 |
| 3 | Before .. | .. | .. | .. | .. | .. | 14.4 | 17.1 |
| | After .. | .. | .. | .. | .. | .. | 12.6 | 15.4 |

Allowing for the average effect of the colouring matter, as thus indicated, the average lead number per 1 per cent. of acid shows that about 19 per cent. of the total acid is malic acid. The extreme figure found might indicate up to 35 per cent., but this was obtained in the early days of the lead method. In only one later sample does the proportion seem to be at all high—about 32 per cent.

The pH values are on the whole rather higher (about 0.1) than would correspond to the proportion of combined to total acid, assuming the latter to be citric acid or a mixture of citric and malic acids. Possibly somewhat high results may have been due to the upward drift during the determination, mentioned earlier. This was often rather marked with blackcurrants.

Plums.—The average figure for insoluble solids is higher than previously recorded,¹ but there is little difference in the averages for soluble solids and free acid. An appreciably higher maximum value for the insoluble solids is to be noted.

Few samples were examined by the lead process, as, until the use of the pectin-free solution was introduced in 1932, the titrations with stone fruits were difficult and very probably in error. This may account for the very low figures (per 1 per cent. of acid) obtained for the two samples examined in 1931.

The pH values, when compared with the proportion of combined to total acid, were on the whole rather lower (about 0.1) than would correspond to the neutralisation curves of either malic or citric acid.

Greengages.—The average and maximum soluble solids are much higher than before¹; the same applies to a less extent to the free acid.

Damsons.—A rather lower average than previously was found for the free acid.¹ Extreme figures for insoluble and soluble solids and free acid all lie within the previous limits.

The aqueous lead numbers indicate very little citric acid. The two results of this determination for 1931 samples are open to some doubt for the reason mentioned in discussing the results for plums.

Apples.—No new extreme limits were established for insoluble and soluble solids and free acid, and the averages for these are close to the previous ones.¹

There is some evidence, on comparing the pH values with the notes on flavour and appearance, that the pH followed the degree of ripeness, the riper fruit having higher pH . A correspondence of this kind was not observed with the other fruits.

Bilberries.—The Cumberland sample had lost a little juice in transit, and the figure for insoluble solids may therefore be slightly high. The percentage of the soluble constituents would not have been appreciably altered, however.

Blackberries.—Lower minimum figures than before have been found for the insoluble solids and free acid; also a much higher maximum. The very wide range of free acid content shown by this fruit may be noted. Since the combined acid does not vary correspondingly, the pH tends to follow the free acid and therefore also shows a wide range.

Apricots and Apricot Pulps.—No figures for apricots were included in the earlier paper.¹

Although there is no absolute certainty, a general consideration of the figures for each of the samples of pulp, in comparison with the averages and with the data for the fresh fruit, makes it reasonably likely that but little water had been used in their preparation. It may be mentioned that several other samples were met with in which the analysis pointed fairly definitely to the addition of an appreciable proportion of water. These, of course, have not been included.

The lead numbers per 1 per cent. of acid would, of course, not be affected by any added water. They indicate about 40 to 50 per cent. of the acids to be malic.

The pH values, when compared with the proportion of combined to total acid, lie fairly close to the neutralisation curve of a mixture of equal parts of citric and malic acids.

USE OF THE DATA IN DETERMINING THE FRUIT CONTENT OF JAMS

Of the analytical characteristics included in the Tables, the insoluble solids content has perhaps been the most frequently used in assessing the fruit content of jams, but suffers from the disadvantage of the possible uneven distribution of the fruit from jar to jar. This difficulty does not apply to the soluble constituents of the fruit, since these, as a result of the boiling, are fairly evenly distributed throughout the liquid or jelly portion of the jam, including that part which permeates the fruit itself, and the effect of the uneven distribution of the fibrous portions of the fruit is negligible.

Unfortunately, the total soluble constituents derived from the fruit cannot be determined in a jam, because of the merging of these constituents, part of which are sugars, in the added sugar in the jam. Hence the data for soluble solids content,

although affording a useful and independent figure in the examination of fruit pulps. are useless for jams.

The acid and salt constituents (free and combined acid) are, however, available, but here again certain difficulties arise. In the first place, when a jam is made from sulphited fruit pulp, any oxidised SO_2 in the pulp remains as sulphate in the finished jam, an equivalent amount of free fruit acid being liberated from the salt constituents. Hence the free acid will be higher and the combined acid lower than in the original fruit. The sum of the two, however, will be unaltered. Under such conditions, therefore, only the total fruit acid in the jam can be accepted as being directly related to that in the original fruit.

A second difficulty arises from the possible presence in the jam of added liquid pectin containing a certain amount of acid derived from the apple material used as a source, or added for the purpose of extracting the pectin. The lead precipitation method² was introduced to overcome this difficulty, and gives useful results with the citric acid fruits.

Another source of uncertainty is the possible addition, during the boiling of the jam, of some acid or salt such as citric acid or a citrate. The magnitude of this difficulty, however, should not be exaggerated. It is very unlikely that any such additions would amount to more than the equivalent of a few per cent. of fruit in the jam; otherwise the effects on flavour, colour, or setting properties would be likely to be objectionable.

INTERPRETING RESULTS.—The discussion of these points indicates that the determination of the fruit content of a jam is beset with difficulties, and, in general, the results are only approximations unless the composition of the fruit used in making the jam is known. It should be noted, however, that the lower the fruit content the closer the results will be to the truth (in terms of percentage of fruit in the jam).

As stated in a previous communication,¹ it is unwise to rely on the insoluble solids figure alone, especially when this is obtained from only one sample. With the best will in the world it is not possible for the manufacturer to ensure that the fibre of the fruit is uniformly distributed throughout all the jars from a single batch. Public Analysts meet with a serious difficulty in this respect, for, should it be found desirable to initiate a prosecution under the Sale of Food and Drugs Act, they must confine their report to the official sample. On the other hand, it has been a growing practice of many authorities of late years to take informal samples, and while the results of these cannot be referred to in a prosecution, the knowledge obtained from their analyses should strengthen the analyst's conclusions. If the analyses of two or more samples of the same make of jam are compared, it will generally be found that, while the insoluble solids may vary considerably, the total acidity and other constituents are reasonably uniform. When this happens it would obviously be unfair to base an opinion on the insoluble solids figure obtained in the analysis of the official sample, if this happened to be lower than the results obtained from the informal samples.

If all the determinations suggested below are carried out, a much sounder conclusion can be arrived at from a consideration of all the figures for fruits containing mainly citric acid. It is true that the lead numbers are much less helpful with fruits containing mainly malic acid, *e.g.* plums, apples, etc., but generally these fruits are so cheap that there is little inducement to lower the quality of the jam seriously.

In calculating the fruit content of a jam from all the analytical data, both the minimum and average figures for the particular fruit should be used. Should the results calculated from the lead numbers and acid figures on the minimal basis indicate a deficiency in fruit, the analyst would be justified in initiating proceedings. It would not, however, be safe to accept the results based on a similar calculation from the insoluble solids figure, unless they were confirmed by results obtained from

other samples or by those calculated from the acid figures and lead numbers on the basis of average data.

When a low fruit content is indicated by the calculation of all constituents on the basis of the average data for these constituents, it is suggested that no prosecutions should be initiated, but that the manufacturers should be given an opportunity of proving that the full quantity of fruit was used. These instances occur as the result of more than the normal amount of condensed steam entering the pulp. Steam pulps must always contain condensate. It should be noted in this connection that fruit preserved with sulphur dioxide sometimes contains excess of water, and the manufacturer may be unaware of this fact. This occurs not infrequently with blackcurrant pulp which is steamed before preservation, and arises from an excessive condensation of steam. Without a full analysis of the contents of each cask, it is difficult to detect occasional casks of such pulp. Manufacturers have been warned to buy pulp made by the hot process on the basis of weight of fresh fruit per cask, and to check this by determinations of the acidity and specific gravity of the juice.

It should be noted that, so far, no single sample of fruit has been found to contain the minimum percentages of both the insoluble solids and acids. With the raspberries containing the lowest percentage of insoluble solids, the acids were up to the average. Blackcurrants having a low insoluble solids have been found to contain less acid than the average, but always more than the minimum. Therefore when considering the results it should not be necessary to base a decision on the results calculated from all constituents on the minimum basis.

A microscopical examination of the jam should always be made, particularly of jams containing seeds, *e.g.* raspberry jam. Cases have been known in which seeds had been added in substantial amount to a raspberry and gooseberry jam, but the very low proportion of raspberry fibre, other than seed, revealed the sophistication, which was afterwards admitted. The proportion of seeds to fibre is also a valuable indication of this form of adulteration. Some results are given in the tables indicating the proportions generally occurring in the original fruit.

IMPROVEMENTS IN THE LEAD PRECIPITATION METHOD AS APPLIED TO JAMS

In applying the lead method to jams, as described in the original paper,² experience has revealed some minor difficulties and sources of uncertainty. These and the means of dealing with them are described below.

FILTRATION OF JAM SOLUTION.—Filtration of the 50 per cent. extract of the jam through paper is often slow. This can be avoided by using coarse muslin or a 30-mesh sieve.

POSSIBLE ERROR IN DETERMINATION OF COMBINED ACID.—Errors in the determination of the combined acid have been found to arise owing to traces of sulphates or chlorides in the jam. These substances, when ignited at even a moderate temperature in presence of an excess of sugar, decompose and form carbonates, thus increasing the alkalinity of the ash and causing the combined acid to appear high. If sugar is absent, or is present only in relatively small proportion as in the fruits themselves, this decomposition does not occur, at least with small amounts of these salts.

It was found that the effect could be prevented by adding a sufficient amount of alkaline material, such as potassium or calcium carbonate, to the solution before ashing. For 50 ml. of pectin-free filtrate, 30 ml. of *N*/10 potassium carbonate solution is a suitable addition. In dissolving the ash subsequently, of course, more acid is required, and in calculating the alkalinity of the ash, that of the added carbonate must be deducted.

AVOIDANCE OF ERRORS IN THE PRECIPITATION OF THE LEAD MALATE.—It was shown in the original paper² that the proportion of lead malate which is precipitated varies somewhat with the amount present, and with the amount of citrate also present.

The proportions of the two acids must be kept within certain rough limits in order to secure precipitation of a constant proportion of the malate.

This was provided for in analysing different fruits by arranging for the amount of fruit extract taken for the analysis to be varied, and for suitable amounts of either malic acid or citric acid to be added, according to the kind of fruit.

These arrangements may sometimes be interfered with in determining the aqueous lead number of jams containing much added pectin. The presence of the latter entails that a larger proportion of malic acid is present than in the fruit from which the jam is named. Some of the acid is also perhaps lactic acid. This means that the jam from a fruit which is naturally a "citric acid fruit" (Group ii) comes to resemble, in its acid content, a "mixed acid fruit" (Group i); and a jam from a "mixed acid fruit" (Group i) approaches a "malic acid fruit" (Group iii). There will be a tendency for the lead malate to be precipitated to a smaller extent than under the standard conditions. This is most likely to occur with gooseberry, apricot or blackberry jams containing much liquid pectin.

The difficulty can be overcome by treating the jam in question according to the group it tends to resemble. Thus for the aqueous lead number of an apricot jam of this type, the amount of pectin-free filtrate to be taken will be that containing about 0.65 g. of total acid; and instead of 3 ml. of 10 per cent. malic acid solution, 3 ml. of 5 per cent. citric acid solution will be added. Usually it is known beforehand that the sample is one likely to require this modification. If this is only discovered after the analysis has been carried out in the usual way, the determination of the aqueous lead number should be repeated as modified.

The acetone lead number does not appear to be liable to this source of error.

INSUFFICIENCY OF SOLUTION FOR TITRATIONS.—It sometimes happens in analysing jams very low in acid content, that the amount of pectin-free filtrate available is insufficient to permit of using the proper amount for the aqueous lead number. In that event the deficiency may be made up with a definite amount of citric or malic acid, as required. An allowance for this can be made, though the accuracy of the final lead number is reduced to some extent.

An alternative method is to take two-fifths quantities for the lead number determinations, the amounts of reagents, of course, being reduced in the same proportion, and to make up the mixture to 100 ml. before filtration. For the first rough titration, 25 ml. of the filtrate should be taken, the full 50 ml. being used for the final titration.

ADDITION OF MALIC OR CITRIC ACID TO CONTROL THE PRECIPITATION OF LEAD MALATE.—It has been pointed out that there is a lack of clarity in the requirement that the strength of the solutions of these acids should be correct to within 1 per cent. of the total.² This statement means that the strength of the solutions should be within the range 9.9 to 10.1 per cent. or 4.95 to 5.05 per cent. respectively, and does not refer to the total acid in the solution taken for the test.

IMPROVEMENT IN END-POINT OF THE LEAD TITRATION.—This has already been described in connection with the analysis of the fruits (see p. 542).

CORRECTION OF LEAD NUMBERS FOR SULPHATE PRESENT IN JAMS.—Sulphate may be present in jams as a natural constituent of the fruit or as a result of oxidation of sulphur dioxide either in pulp used in making the jam, or, to a negligible extent, in the jam itself. Lead sulphate is partly insoluble under the conditions of the aqueous lead precipitation and completely so under those of the acetone lead precipitation. In the former, neither the amount precipitated nor the amount retained in solution is constant; both vary with the amount of sulphate present in the mixture. This effect is seen in the following experiment, in which different amounts of sulphate were added to solutions prepared to represent jam solutions under examination by the aqueous lead test.

Mixtures as shown (Table XVIII) were dissolved in water, and each was treated with 20 ml. of 10 per cent. lead acetate solution, made up to 250 ml.,

mixed, and filtered. The titrations with molybdate were carried out as usual, on 50 ml. of filtrate, two individual workers each making two titrations. The averages of the four titrations of each filtrate are shown in the Table.

From these figures it is possible to derive corrections to be applied to the titration of the aqueous lead number. These are shown in Table XIX. The determination of the sulphate can be made on the ash if the precaution is taken of adding excess of alkali prior to ashing, as already described (p. 556).

TABLE XVIII—LEAD PRECIPITATED IN THE AQUEOUS LEAD TEST BY VARYING AMOUNTS OF SULPHATE

(7.5 ml. of 5 per cent. citric acid, 10 ml. of *N*/10 NaOH and 50 g. of sugar in each mixture)

| Potassium sulphate added g. | Back titration with molybdate ml. | Difference due to extra lead precipitated by sulphate ml. |
|--------------------------------|---|---|
| 0.0 | 9.81 | — |
| 0.0125 | 9.66 | 0.15 |
| 0.025 | 9.60 | 0.21 |
| 0.050 | 9.41 | 0.40 |
| 0.075 | 9.30 | 0.51 |

The acetone lead number is also raised by any sulphate present. Here the lead sulphate is completely precipitated, and the correction is therefore equal to the lead equivalent of the sulphate present. Each 0.01 g. of K_2SO_4 in the amount (P) of pectin-free filtrate used for the test requires a deduction of 0.25 ml. from the titration difference before the acetone lead number is calculated.

TABLE XIX—CORRECTION FOR SULPHATE (AQUEOUS LEAD NUMBER)

| Sulphate as K_2SO_4 in the amount (P) of pectin-free filtrate taken g. | Deduction from titration difference ml. |
|--|---|
| 0.01 | 0.1 |
| 0.02 | 0.2 |
| 0.03 | 0.3 |
| 0.05 | 0.4 |
| 0.07 | 0.5 |
| 0.10 | 0.6 |

It may be mentioned that the amounts of sulphate in jam only become significant in this respect when fruit pulp containing much oxidised sulphur dioxide or sulphite has been used in manufacture.

MODIFIED CALCULATION OF THE FRUIT CONTENT FROM THE LEAD NUMBERS.—In the earlier paper² the acetone lead number (per 1 per cent. of total acid) in a jam containing no lactic acid was taken to be 15, the value for citric or malic acid. On this figure was based the calculation of, and the allowance for, any lactic acid which might be present.

The data now reported show that the value is usually slightly higher than 15, and appreciably higher with black- and redcurrants. For these fruits it was shown that the high figures appear to be due to the colouring matters. These colouring matters are not removed in the ordinary way, and, as in any event they are natural constituents of the fruit, it seems more correct in calculating the fruit content of jams to use the average value for the acetone lead number of the particular fruit in question.

For blackcurrants which, with redcurrants, differ most widely from the figure for the pure acids, a calculation shows that the fruit content indicated will be about 2 per cent. too low if the figure 15 is used in place of the average figure 16.25 found for that fruit. The percentage of lactic acid calculated from the acetone lead number is likely to be more nearly correct if a value appropriate to the kind of fruit present is used.

For convenience in reference, the average values for the lead numbers (per 1 per cent. of acid) of the fruits are collected in Table XX, and the average total acid contents are also shown. The values of L_1 and A_1 are those to be substituted in formula VI of the earlier paper²; and the values of L_1' are those now to be used in

TABLE XX—AVERAGE VALUES FOR THE TOTAL ACID CONTENT AND LEAD NUMBERS (PER 1 PER CENT. OF ACID) OF JAM FRUITS

| | Total acid (A_1) Per Cent. | Aqueous lead number per 1 per cent. of total acid (L_1) | Acetone lead number per 1 per cent. of total acid (L_1') |
|---------------------|--------------------------------------|--|---|
| Gooseberries | 2.37 | 10.36 | 15.4 |
| Strawberries | 1.31 | 12.68 | 15.31 |
| Raspberries | 2.07 | 13.36 | 15.34 |
| Redcurrants | 2.87 | 13.46 | 16.25 |
| Blackcurrants | 3.86 | 13.47 | 16.25 |
| Blackberries | 1.51 | 9.22 | 15.4 |
| Apricots | 2.20 | 10.6 | 14.9 |

place of the citric or malic acid value of 15 in forming an opinion as to the presence of lactic acid and in calculating and correcting for it. It should be noted that if L' , the acetone lead number per 1 per cent. of acid, corrected for sulphate as described above, lies above the value of L_1' for the fruit in question, there can be no allowance for lactic acid to be made in evaluating l (aqueous lead number per 1 per cent. of acid), since obviously there cannot be a negative amount of lactic acid. The fact that L' lies above the average value for the fruit simply means that the fruit used is slightly divergent from the normal. There may actually be a small amount of lactic acid present, but in such a case it is clearly impossible to allow for it. The error is not likely, however, to be significant.

For routine tests, the titration of the ash to arrive at the combined acid (and hence the total acid) may sometimes be omitted, reliance being placed simply on the acetone lead number for finding the total fruit acid (*i.e.* less lactic acid). The corrected value of a for substitution in formula VI² is then given directly by

$$a = \left(\frac{L'}{L_1'} \right)$$

In such a shortened procedure, in order to find the amount of pectin-free filtrate to be used for the lead number determinations, it is necessary to make a guess at the approximate total acid content from the free acid titration alone.

In conclusion, we wish to thank Messrs. Chivers & Sons Ltd., for help in obtaining samples of fresh fruits from Holland, and the Council of the Research Association for permission to publish this work with a view to assisting in the maintenance of the Jam Standards.

REFERENCES

1. T. Macara, *ANALYST*, 1931, **56**, 35.
2. C. L. Hinton, *id.*, 1934, **59**, 248.
3. T. Macara, *id.*, 1931, **56**, 391.
4. —, *id.*, 1935, **60**, 592.

BRITISH ASSOCIATION OF RESEARCH FOR THE COCOA,
CHOCOLATE, SUGAR CONFECTIONERY AND JAM TRADES
HOLLOWAY, LONDON, N.7

May 15th, 1940

The Gravimetric Determination of Phosphate and Vanadate

By DAVID T. LEWIS, PH.D., A.I.C.

THE possibility of determining various acid radicals by precipitation as insoluble uranyl salts deserves consideration, as the high atomic weight of uranium makes the percentage weight of the radical in the precipitate very small and thus considerably increases the accuracy of the determination. Lewis and Davis¹ have described a method for determining the arsenate radical based on the precipitation of uranyl ammonium arsenate. The present paper deals with the conditions of precipitation, analysis and use of the corresponding salts of phosphoric and vanadic acids.

DETERMINATION OF PHOSPHATE GROUP.—Uranyl ammonium phosphate was first described by Arendt and Knop,² and L. Barthe³ has shown that the alkyl amines yield similar derivatives. Priority in suggesting the use of this precipitate in quantitative analysis must be given to Kitchin,⁴ who studied the precipitation of disodium hydrogen phosphate with uranyl salts. Although Kitchin employed the formula $(U_2O_3)_2P_2O_5$, then in vogue for uranyl phosphate, and accepted the rough atomic weight value $U = 120$, yet his results on recalculation in the light of modern data show that he achieved an accuracy lying within 0.1 per cent.

I have studied the application of this method to the phosphates of sodium, ammonium, calcium and barium, and have also used it for the determination of phosphorus in urine and organic compounds. The results indicated that it affords a rapid and accurate method for the determination of phosphate. The general procedure for organic compounds is briefly as follows:—

Determination of Phosphate.—The organic compound (0.1 to 0.4 g.) is decomposed in a Kjeldahl flask by the method of Lewis and Davis (*loc. cit.*)¹ or by means of a mixture in equal volumes of fuming nitric acid and sulphuric acid. A known volume of urine may be similarly decomposed. After complete oxidation, the colourless liquid is neutralised with ammonia (methyl red as indicator) and then slightly acidified with acetic acid, and treated with 2 g. of ammonium acetate. The solution is heated to boiling, approximately $N/10$ uranyl acetate solution is added dropwise until precipitation is complete, and, after (preferably) standing overnight, the precipitate is transferred to a No. 40 Whatman filter-paper, dried, and ignited in a platinum crucible to uranyl pyrophosphate:— $2(UO_2)NH_4PO_4 \rightarrow (UO_2)_2P_2O_7 + 2NH_3 + H_2O$. 1 g. of pyrophosphate $\equiv 0.1988$ g. of P_2O_5 . Overheating will make the residue green, owing to slight decomposition, but addition of a few drops of nitric acid and gentle re-ignition gives the stable yellow pyrophosphate.

The results with organic compounds were very satisfactory. In the inorganic work it was found that the alkali metal and barium salts gave practically theoretical results, but in presence of much calcium the results were generally from 0.1 to 0.5 per cent. too low.

URANYL AMMONIUM VANADATE.—This salt does not appear to have been described (*cf.* Mellor, "*Treatise of Inorganic and Theoretical Chemistry*," 1929, Vols. 8, 9 and 12), although J. J. Berzelius obtained uranium metavanadate from alkali tetravanadate solutions. Lachatre⁵ has shown that ammonium metavanadate in acetic acid solutions gives the tetra- and hexavanadates.

It has been found that, under definite experimental conditions, it is possible to precipitate the vanadate quantitatively as the uranyl ammonium salt. Analysis of the dried yellow precipitate gave NH_3 , 3.90; U , 55.95; loss on ignition, 12.1 per cent.; this corresponds with the formula $NH_4(UO_2)VO_4 \cdot 3/2H_2O$, which requires NH_3 , 3.95, U , 55.35, and ignition loss, 12.32 per cent. Friedel and Cumenge⁶ have carried

out an accurate investigation of the analogous mineral carnotite and suggest the similar formula, $K(UO_2)_2VO_4 \cdot 3/2H_2O$.

Experimental Method.—The following method was applied to pure (AnalaR) ammonium metavanadate dried in a vacuum desiccator, and dissolved in dilute sulphuric acid:—To 25 ml. of the vanadate solution, containing about 0.05 g. of V_2O_5 , ammonium hydroxide is added until the solution is just alkaline (neutral red, light filtered with methylene blue).^{*} The solution is then made very slightly acid with acetic acid and 2 g. of ammonium acetate are added. The solution is heated to boiling and, on addition of excess uranyl acetate, a dense, yellow precipitate is thrown down. After standing for some hours to facilitate the formation of definite crystals, this precipitate is transferred to a No. 4 Jena glass Gooch crucible and dried in an air oven at 105° C. to constant weight. 1 g. of $NH_4(UO_2)_2VO_4 \cdot 3/2H_2O \equiv 0.2114$ g. of V_2O_5 . Some typical analyses corroborating this factor are given below. It is important to adjust the pH of the solution fairly carefully, as the vanadate precipitate is far more soluble than the corresponding phosphate and arsenate.

| NH_4VO_3 g. | A. V_2O_5 g. | B. $NH_4(UO_2)_2VO_4 \cdot 3/2 H_2O$ g. | A/B Per Cent. |
|------------------|----------------------|---|------------------|
| 0.07382 | 0.05738 | 0.2736 | 20.97 |
| 0.08576 | 0.06667 | 0.3160 | 21.16 |
| 0.06536 | 0.05080 | 0.2409 | 21.09 |
| 0.1001 | 0.07784 | 0.3599 | 21.50 |
| 0.04802 | 0.03733 | 0.1782 | 20.96 |
| 0.0096 | 0.00746 | 0.0360 | 20.64 |

Mean = 21.05

Concentrations of V_2O_5 exceeding 0.08 g. in 25 ml. are to be avoided, as the precipitate is liable to contamination by adsorbed impurities; with concentrations in the above range, however, the results are very satisfactory, and, owing to the heavy nature of the precipitate formed, the method can be employed for very dilute vanadate solutions. Attempts were made to ignite the precipitate to uranyl pyrovanadate $(UO_2)_2V_2O_7$, after filtration, but, although the results were fairly good (within 0.5 per cent.), they were not sufficiently exact for accurate work, and the method should be employed only when an approximate result is required. It is probable that the decomposition by heat is attended by some slight by-reaction which makes the ignition procedure rather inexact; the Gooch method has been found to be far superior.

REFERENCES

1. D. T. Lewis and V. E. Davis, *J. Chem. Soc.*, 1939, 285.
2. R. Arendt and W. Knop, *J. Prakt. Chem.*, 1857, 70, 385.
3. L. Barthe, *Compt. rend.*, 1911, 152, 1396.
4. A. Kitchin, *Chem. News*, 1873, 27, 199.
5. M. Lachartre, "Mellor's Treatise of Inorganic and Theoretical Chemistry," 1929, Vol. 9, 759.
6. C. Friedel and E. Cumenge, *Compt. rend.*, 1899, 128, 532.

TATEM CHEMISTRY LABORATORIES,
UNIVERSITY COLLEGE, CARDIFF

June, 1940

^{*} A 0.1 per cent. alcoholic solution of neutral red mixed with an equal volume of a 0.1 per cent. solution of methylene blue. For details of light filtering, see Vogel, "Textbook of Quantitative Analysis," p. 69; Fowles, "Volumetric Analysis," p. 71.

Studies in Internal Electrolysis. V

The Determination of Small Quantities of Copper in Presence of Cadmium

By JAMES G. FIFE, M.Sc., F.I.C.

THE separation and determination of copper and cadmium has recently aroused considerable interest, as is shown by the work of Rây and Bose,¹ Shennan,² Majumdar³ and Smith.⁴ The object of the present investigation was to provide a rapid electrolytic method for the determination of small quantities of copper in presence of cadmium. It was found that this could be effected by the method of internal electrolysis.

TABLE I

| Experiment | Copper taken g. | Conc. sulphuric acid added ml. | Copper found g. | Time of electrolysis, minutes |
|------------|--------------------|---|--------------------|-------------------------------------|
| 1 | 0.0020 | nil | 0.0020 | 20 |
| 2 | nil | nil | nil | 17 |
| 3 | 0.0040 | nil | 0.0040 | 30 |
| 4 | 0.0010 | nil | 0.0011 | 14 |
| 5 | 0.0005 | nil | 0.0006 | 21 |
| 6 | 0.0080 | nil | 0.0080 | 24 |
| 7 | 0.0100 | 2 | 0.0102 | 34 |
| 8 | 0.0025 | 2 | 0.0025 | 25 |
| 9 | 0.0022 | 1 | 0.0022 | 18 |
| 10 | 0.0007 | 2 | 0.0007 | 18 |
| 11 | 0.0150 | 3 | 0.0152 | 36 |
| 12 | 0.0030 | 3 | 0.0030 | 30 |
| 13 | 0.0200 | 3 | 0.0201 | 50 |
| 14 | 0.0004 | 4 | 0.0004 | 19 |

The apparatus employed was that described by Sand⁵ with the modification used in the determination of small proportions of cadmium and of nickel in zinc.⁶ The anodes consisted of cadmium tape prepared by rolling out pure stick cadmium and soldering copper connecting wires to the top end of the cadmium tape. Satisfactory results have been obtained by using a platinum cathode, an anolyte containing cadmium sulphate equivalent to 5 g. of cadmium and also 0.2 g. of hydroxylamine sulphate per 100 ml., and a catholyte of approximately 300 ml. containing the copper to be determined (added as sulphate), 0.2 g. of hydroxylamine sulphate, cadmium sulphate equivalent to 5 g. of metal and in some experiments small quantities of sulphuric acid. The electrolysis was carried out between 70 and 80° C., for about 15 to 60 minutes, according to the amount of copper present; when a new apparatus is set up, however, the time required for solutions of known composition should be determined.

The results are shown in Table I.

TABLE II

| Experiment | Copper taken g. | Cadmium added g. | Copper found g. | Time of electrolysis, minutes |
|------------|--------------------|---------------------|--------------------|-------------------------------------|
| 15 | 0.0096 | 5.17 | 0.0095 | 51 |
| 16 | 0.0117 | 5.35 | 0.0117 | 58 |

In further experiments, known amounts of pure cadmium and electrolytic copper were dissolved in 10 ml. of conc. sulphuric acid and 2 ml. of conc. nitric acid, the solution was neutralised with ammonia and then acidified with 3 ml. of conc.

sulphuric acid, and 0.2 g. of hydroxylamine sulphate was added. The solution was electrolysed as described above. The results are shown in Table II.

In Expt. 16 the determination was carried to constant weight, showing that prolongation of the electrolysis has no harmful effects.

It is known that copper and cadmium form a eutectic mixture containing approximately 1.2 per cent. of copper.⁷ A copper-cadmium alloy was prepared containing 1.03 per cent. of copper, and weighed portions of this were dissolved in 10 ml. of conc. sulphuric acid and 2 ml. of conc. nitric acid. The solution was filtered, neutralised with ammonia and acidified with 3 ml. of conc. sulphuric acid, and 0.2 g. of hydroxylamine sulphate was added.

The results are shown in Table III.

TABLE III

| Wt. of alloy taken g. | Copper found g. | Copper Per Cent. |
|--------------------------|--------------------|---------------------|
| 0.436 | 0.0044 | 1.01 |
| 1.518 | 0.0158 | 1.04 |

I wish to thank Dr. A. J. Lindsey for his interest in this work.

REFERENCES

1. P. Rây and M. K. Bosc, *Z. anal. Chem.*, 1933, **95**, 400.
2. R. J. Shennan, *ANALYST*, 1939, **64**, 14.
3. A. K. Majumdar, *id.*, 1939, **64**, 874.
4. G. S. Smith, *id.*, 1940, **65**, 87.
5. H. J. S. Sand, *id.*, 1930, **55**, 309.
6. J. G. Fife, *id.*, 1936, **61**, 681.
7. H. Neville, *J. Chem. Soc.*, 1892, **61**, 898.

THE SIR JOHN CASS TECHNICAL INSTITUTE,
JEWRY STREET, LONDON, E.C.3

July 16th, 1940

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

A LABORATORY TEXTILE SCOURING APPARATUS

METHODS of removing, by scouring, oils used for textile fibre lubrication, have been studied, and detergent baths used for scouring have been compared. Many of these methods proved unsatisfactory, but the simple apparatus shown in the diagram has given results which are as consistent as those obtained with the Society of Dyers and Colourists' Wash Wheel.¹

It consists of a flask F containing the detergent solution, in which the material to be scoured is placed. The exit tube E has a Gooch crucible G fixed to the end to act as a filter. The inlet tube I has a Bunsen non-return valve at the end.

The detergent solution is circulated through the reciprocating pump R. This consists of a cork collar C hollowed out to form a seating for a steel ball B, thus forming a non-return valve. The piston of the pump has a 1-inch stroke and consists of a cork P, bound round with thin string to form "piston rings." The piston rod is a glass tube connected by means of a connecting rod to the eccentric EX, which makes 8 revolutions per minute.

When the pump is in operation the scouring liquor is forced intermittently in one direction only (shown by the arrows) and in the flask F a swirling motion is set up, which agitates the material being scoured.

The whole apparatus is contained in a water-bath controlled by the thermostat T.

An example is given in Fig. 2, to illustrate the reproducibility obtainable. Wool was oiled under standard conditions with three oils adjusted to have the same viscosity, viz. S = sperm oil,

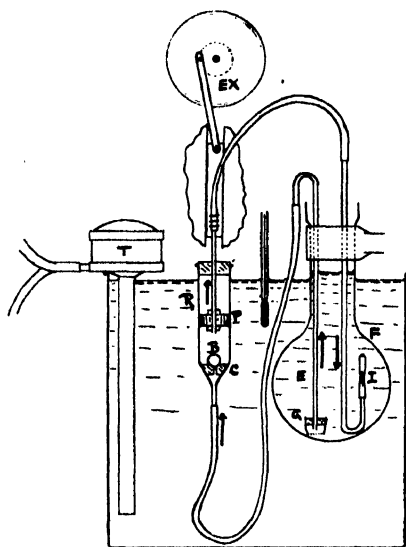


Fig. 1

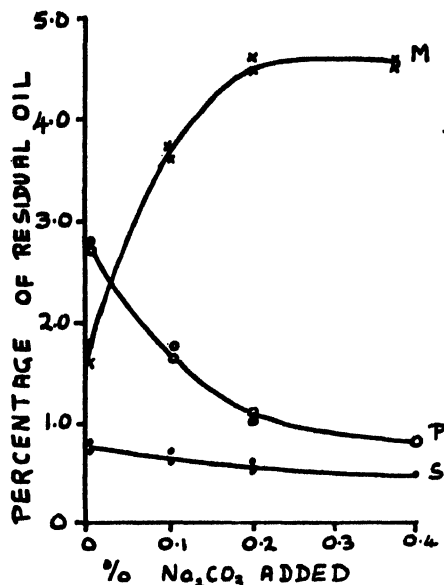


Fig. 2

M = refined mineral oil, and P = refined mineral oil containing a polar compound (6.3 per cent. of oleyl alcohol, as first proposed by J. B. Speakman). Scouring was carried out in every instance in a 0.2 per cent. soap solution for 20 minutes at 37° C., the amount of soda being varied.

REFERENCE

1. *Society of Dyers & Colorists: Report of Fastness Committee.*
 LISTER & Co., LTD.
 MANNINGHAM MILLS
 BRADFORD

W. GARNER

April 18th, 1940

DETECTION OF SMALL AMOUNTS OF PROTEINS ON THE SURFACE OF METAL PLATES

DURING the past two years considerable use has been made in these laboratories of the chloramine test for proteins¹ for the detection of small amounts of protein material—usually albumen or casein—on the surface of zinc or aluminium lithographic printing plates. When investigating the causes of various defects in lithographic plates it has often been important to ascertain whether black specks and patches on the plate are, or are not, printing images formed by albumen or casein.

The chloramine test was originally devised by Cross and Bevan for the detection of small amounts of gelatin sizing in paper and for showing localised spots of gelatin. The test has been modified slightly for use with metal plates and is now carried out in these laboratories in the following manner:

A small portion of the plate to be tested is cut out and all printing ink is cleaned off by thorough rubbing with cotton wool and benzene. The cleaned plate is then placed in a test-tube and moist chlorine gas is passed in for half-an-hour to chlorinate the protein. (This is more convenient than Cross and Bevan's original method of chlorination by immersion in hypochlorous acid solution, which is liable to attack the metal plate.) The strip is removed, rinsed with cold water and placed in a 2 per cent. solution of sodium dihydrogen phosphate at 45° to 50° C. for exactly five minutes, to destroy free chlorine and hypochlorous acid, after which it is transferred to a solution containing 1 per cent. of potassium iodide and 0.1 per cent. of starch, or the solution may be poured on to the plate; specks of protein matter on the surface are stained with the dark blue starch-iodine colour.

The test has been found to work well with specks of light-hardened bichromated albumen and with specks of casein on a plate. It has also been found possible in several instances to determine whether black specks ("scumming") on a litho plate were, or were not, due to specks of

the albumen used in the preparation of the plate not having been washed off properly. Another application of the test has been to ascertain whether scumming has arisen from casein transferred to the plate from the coating of the paper being printed.

I wish to thank the Council of the Printing and Allied Trades Research Association for permission to publish this note.

REFERENCE

1. C. F. Cross, E. J. Bevan and J. F. Briggs, *J. Soc. Chem. Ind.*, 1908, **27**, 260. C. F. Cross and E. J. Bevan, "*A Text Book of Paper-Making*," London, 1936, p. 420.

J. H. YOUNG

10 ROBIN HOOD COURT
SHOE LANE
LONDON, E.C.4

August 8th, 1940

Notes from the Reports of Public Analysts

The Editor would be glad to receive the Annual or other Reports of Public Analysts containing matter of special interest to the Society. Notes made from such Reports are submitted to the Publication Committee.

CITY OF BIRMINGHAM

REPORT OF THE CITY ANALYST FOR THE SECOND QUARTER, 1940

OF the 1388 samples submitted, 23 were taken formally.

MILK CHEESE.—A sample sold as "milk cheese" was found to have been made from partly skimmed milk instead of whole milk. The manufacturers agreed to state the facts on the label.

"VINEGAR ESSENCE."—A sample consisted of a colourless, dilute solution of acetic acid of 19.4 per cent. strength, and was labelled "Vinegar Essence (Kosher) 20 per cent. Five parts of water to 1 part of Essence." If these instructions had been followed, the strength of the resultant mixture would have been only 3.25 per cent. It was found that the firm selling this product had formerly been conducted by two refugees, and that since their internment the business had been disorganised. The sale was discontinued.

SACCHARATED IRON TABLETS.—A sample contained only 2.3 g. of saccharated iron carbonate per tablet instead of 4.9 g., the remainder being in the oxidised condition. The deficiency may have been due to deterioration during storage. The firm was cautioned.

COLOURLESS TINCTURE OF IODINE.—A sample contained 7.1 per cent. of free ammonia instead of 1.4 per cent. and 4.6 per cent. of ammonium iodide instead of 3.25 per cent. A formal sample from the same shop contained 2.3 per cent. of free ammonia and only 1.45 per cent. of ammonium iodide. The vendor attributed the incorrect composition to an error in calculation. He was cautioned.

H. H. BAGNALL

Legal Note

Under this heading will be published notes on cases in which points of special legal or chemical interest arise. The Editor would be glad to receive particulars of such cases.

FALSE TRADE DESCRIPTION OF BLEACHING POWDER

ON August 21st, at Newport Pagnell Police Court, a Cheshire chemical manufacturer was summoned under Section 2 of the Merchandise Marks Act, 1887, for selling bleaching powder under a false trade description—"Chloride of lime." A packet of his preparation, purchased from the Newport Pagnell branch of a chain stores, bore on its label a statement that it was suitable for A.R.P. work. Analysis showed the powder to contain only 13.76 per cent. of available chlorine and to be adulterated with 32 per cent. of sand. It was contended for the defence that the powder could be correctly described as "chlorinated lime" and that that term was synonymous with "chloride of lime." The Bench considered, however, that the description "chloride of lime" implied that the substance was bleaching powder and should therefore comply with the B.P. requirements as to chlorine-content and freedom from "an unusual impurity." According to the B.P. Codex bleaching powder is sometimes known as "chloride of lime."

The defendant was fined £20 with 10 guineas costs.

Ministry of Health

A COMMUNICATION from the Ministry of Health dated September 9th, 1940, notifies the following amendments to the list of Public Analysts appointed by Local Authorities with the approval of the Minister.

| Authority | Analyst |
|---------------------------------------|-------------------------|
| Swinton and Pendlebury, Borough | T. R. HODGSON. |
| West Suffolk, County | W. F. GREEVES (Deputy). |
| Norfolk, County | " " |
| Lowestoft, Borough | " " |
| East Suffolk, County | " " |
| Great Yarmouth, Borough | " " |
| Oxford, County Borough | J. H. WEBER (Deputy). |
| Wakefield, County Borough | F. W. JAFFE (Deputy). |
| Kingston upon Hull, County Borough .. | D. J. T. BAGNALL. |

Ministry of Food

Emergency Powers (Defence)

Food (Jam)

The Jam (Maximum Prices) Order, 1940. Dated August 20, 1940*

THIS Order comprises 8 Articles and 3 Schedules. Article 1 prohibits the sale or exposure for sale of jam of a description mentioned in the first column of par. 5 of the First Schedule unless,

(a) such jam conforms to one or other of the quality standards mentioned in paragraphs 2, 3 and 4 of that Schedule, and

(b) is labelled in accordance with the provisions of the Second Schedule.

Article 2 prohibits the sale by retail of any jam of a description and quality standard mentioned in the Third Schedule at a price exceeding that specified. This provision does not apply to jam sold by retail by the manufacturer thereof, where the total quantity of jam manufactured in the year ended June 30, 1939, did not exceed 5 tons.

Article 3 provides

(1) That in any prosecution for the sale of jam of a quality standard other than that indicated on the label, or of weight less than was represented to the purchaser, it shall be a sufficient defence for the person charged to prove:

(a) that the jam was purchased in the container in which he sold it and with a written warranty as to its quality standard or as to its weight;

(b) that he had no reason to believe that the quality standard or weight was otherwise than as warranted.

(2) A warranty shall only be a defence if the defendant has within seven days of the service of the summons sent to the prosecutor a copy of the warranty with a notice stating that he intends to rely on it, and specifying the name and address of the person from whom he received it, and has also sent a like notice of his intention to that person.

(3) A servant of the person who purchased the jam shall be entitled to rely upon the warranty.

(4) The person by whom the warranty is alleged to have been given shall be entitled to appear at the hearing and to give evidence.

(5) Any statement relating to quality standard or weight on any label affixed to the wrapper or container of the jam or in an invoice or similar document relating to any jam mentioned in the invoice or document shall be deemed to be a written warranty.

Article 4 provides that the Order shall not apply to (a) jam manufactured outside the United Kingdom; (b) the sale of jam sold as part of a meal by a caterer in the ordinary course of his catering business.

Article 5 prohibits any fictitious or artificial transactions or unreasonable charges in connection with the sale or disposition of any jam.

Article 6 authorises the Minister to give or grant general directions, authorisations or licences.

Article 7 provides that infringements of the Order are offences against the Defence (General) Regulations, 1939.

Article 8 provides that the Order shall come into force on September 1st, 1940, and may be cited as the Jam (Maximum Prices) Order, 1940.

* H.M. Stationery Office, York House, Kingsway, London, W.C.2. 1940. Price 2d. net.

THE FIRST SCHEDULE
Provisions Relating to Quality Standards

1. In this Schedule:

- (a) "Fruit Content" means the quantity of fresh fruit or fruit pulp of the variety or varieties of fruit named in the description of the jam (excluding added fruit juice or fruit pectin) used in the manufacture of the jam, expressed as a percentage based on the number of pounds of such fresh fruit or fruit pulp required to be used in making 100 lbs. of finished jam.
- (b) "Minimum percentage of soluble solids" means the minimum percentage by weight of soluble solids ascertained by means of a refractometer when the jam is cold, no correction being made for insoluble solids.
2. "Fresh Fruit Standard Jam" means jam,
 (i) of which the percentage of soluble solids is not less than 68·5 per cent.;
 (ii) of which the fruit content complies with the provisions of the second column of the scale in par. 5 below;
 (iii) of which the fruit content consists of fresh fruit only, and
 (iv) which does not contain any added colouring matter or any added preservative other than sugar.
3. "Full Fruit Standard Jam" means jam,
 (i) of which the percentage of soluble solids is not less than 68·5 per cent., and
 (ii) of which the fruit content complies with the provisions of the second column of the scale in par. 5 below.
4. "Lower Fruit Standard Jam" means jam,
 (i) of which the percentage of soluble solids is not less than 68·5 per cent., and
 (ii) of which the fruit content complies with the provisions of the third column of the scale in par. 5 below.

MINIMUM FRUIT CONTENT

| First column | Second column | Third column |
|-----------------------------------|--|------------------------------------|
| Description of jam | Fresh Fruit Standard Jam and Full Fruit Standard Jam Per Cent. | Lower Fruit Standard Jam Per Cent. |
| Strawberry | 42 | |
| Blackcurrant | 30 | |
| Raspberry | 38 | |
| Green gooseberry | 35 | |
| Red gooseberry | 40 | 20 |
| Victoria plum | 40 | |
| Green or golden plum | 35 | |
| Red plum | 40 | |
| Strawberry and gooseberry | 40(20/20) | 20(10/10) |
| Raspberry and gooseberry | 40(20/20) | 20(10/10) |
| Gooseberry and strawberry | 40(30/10) | 20(15/5) |
| Gooseberry and raspberry | 40(30/10) | 20(15/5) |

NOTE.—The figures in brackets in the second and third column above denote the respective contents of the fruits in the order given.

The SECOND SCHEDULE deals with provisions relating to labelling and the THIRD SCHEDULE gives the maximum retail prices of the three qualities of jam.

=====

STATUTORY RULES AND ORDERS. 1940. No. 606

The Dripping (Maximum Prices) Order, 1940. Dated April 25, 1940*

THIS Order contains eight articles:

(1) "Dripping" means the clear unbleached and unadulterated fat, untreated by any chemical process, of sweet smell, and produced from or by the rendering of Fat and Bones of Sheep, Oxen or Pigs, the finished product of such Rendering or Processing to contain a minimum of 99 per cent. of Saponifiable Matter and a maximum of 2 per cent. of Free Fatty Acids.

"Sale by Wholesale" means any sale other than a sale by retail and includes a sale to a person carrying on business as a fishfryer.

Articles 2 to 5 fix the maximum prices that may be charged and the charge that may be made for a returnable package.

Article 6 provides that the provisions of the Animal Oils and Fats (Provisional Control) (No. 2) Order, 1939(a), as amended(b), relating to Home Melt Technical Tallow shall apply to any Home Melt Edible Tallow which is not dripping as defined in Article 1 of this Order.

* H.M. Stationery Office. 1940. Price 1d. net.

(a) S.R. & O., 1939 (No. 1480), I, p. 1032. (b) S.R. & O., 1940, No. 162.

International Society of Leather Trades Chemists

DETERMINATION OF THE COLOUR OF VEGETABLE TANNIN SOLUTIONS*

THE Committee has studied the application of the Bolton and Williams Colorimeter (ANALYST, 1935, 60, 447) to the measurement of the colour of vegetable tannin solutions and the calculation of the readings into Lovibond figures. In the earlier experiments the calculations (based on a chart by K. A. Williams) were made by means of the formulae:

$$R = 13.32D_3 - 11.98D_2$$

$$Y = 251.0D_3 - 20.30D_2$$

where R and Y are the required Lovibond units, and D_3 and D_2 are "absorption densities" derived from the Bolton and Williams readings at 530 and 640m μ , respectively, by means of equations of the type $D = 2 - \log(100 - \text{per cent. light absorbed}) = K \times \text{depth of layer} \times \text{concentration}$.

A "brightness" figure was obtained by dividing the sum of the red, green and blue (460m μ) transmissions by 3, and the transmissions by subtracting the absorptions from 100.

It was found that with highly coloured liquors there was a pronounced divergence between the two sets of results, and further work showed that the main errors were due to the use of neutral tint Lovibond glasses and to not renewing the copper sulphate solution of the Bolton and Williams instrument often enough. It is recommended that it should be changed at least daily.

In May, 1940, the preparation of a new chart was begun by K. A. Williams, based on more accurate readings than the original ones. A Bolton and Williams colorimeter was adapted to hold the modern Lovibond racks of colour glasses, and it was found that a figure for a combination of two glasses could be obtained, agreeing within the range of the experimental error with the determined figure, by calculation from the separate figures for each glass. From this chart it is anticipated that it will be possible to work out two tables showing the B. & W. figures corresponding respectively with the Lovibond Red and Yellow readings.

The Committee formed the following conclusions:

1. The agreement between the results obtained by different observers with the Bolton and Williams Colorimeter is very good, in view of the fact that most of the observers had never used the instrument before.

2. The concordance between the figures obtained with the colorimeter is very much better than those given by the tintometer.

3. It is agreed that the Bolton and Williams Colorimeter is very simple and easy to work, is very sensitive to differences in colour, and gives definite and reproducible results. The method of reporting results so that they will be easy to visualise is still under consideration, but the new table is very promising.

The Committee records its indebtedness to Mr. Williams for his help and mentions that he has no financial interest in the colorimeter.

International Union of Chemistry

FIFTH REPORT OF THE COMMITTEE ON ATOMS†

THE revised International Table of Stable Isotopes for 1940 is published in English, German, Italian and French, rough or indirect measurements being given in brackets. The following changes in the Table are recommended:

The symbol (M) for Mass Number is replaced by the symbol (A) in conformity with general usage.

LITHIUM.—The figure 7.9 for the percentage abundance of ^6Li is probably too high owing to the uncertain correction for the isotope effect in free evaporation. A lower value (7.5) is now recommended in accordance with the measurements of Hoff Lu (*Phys. Rev.*, 1938, 35, 845).

CARBON.—The very complete investigations of Nier and Gulbransen (*J. Amer. Chem. Soc.*, 1939, 61, 697) show that the percentage abundance of ^{13}C varies appreciably in nature. The mean value 1.1, now recommended, agrees excellently with the earlier work of Brosi and Harkins (*Phys. Rev.*, 1937, 52, 472).

CHROMIUM AND IRON.—Percentage abundances calculated from the work of Nier are recommended (*Phys. Rev.*, 1939, 55, 1143).

MOLYBDENUM.—The more accurate results of Mattauch and Lichtblau (*Z. phys. Chem.*, 1939, B42, 288) are incorporated.

EUROPIUM.—The work of Lichtblau (*Naturwiss.*, 1939, 27, 260) has indicated that the heavier of the twin isotopes of this element is slightly the more abundant.

HAFNIUM.—A new rare isotope has been discovered by Dempster and its abundance estimated to be 0.3 per cent. (*Phys. Rev.*, 1939, 55, 794).

URANIUM.—The measurements of Nier (*Phys. Rev.*, 1939, 55, 150) indicate the presence of the third rare isotope 234 (Uii) and provide accurate figures for the abundances of the other two.

* Report No. 1 of the British Section Committee. *J. Int. Soc. Leather Trades Chem.*, 1940, 24, 257–264.

† Chairman: F. W. Aston. Members: N. Bohr, O. Hahn, W. D. Harkins, F. Joliot, R. S. Mullikin, M. L. Oliphant. 28 Rue St. Dominique, Paris. 1940.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Oxidation Turbidities [in Beer] and their Causes. E. Singruen. (*Modern Brewer*, 1940, 23, 31; *J. Inst. Brewing*, 1940, 46, 285.)—Chill-haze and oxidation-haze are both due to the precipitation of protein-tannin compounds, but the changes which result in the former only are reversible. The latter contains 50 per cent. of protein and a little mineral matter (including silica and iron oxide), and is coloured by pigments and resins. Its formation is controlled by the sensitiveness of the beer to oxidation, which in turn depends on the nature and proportions of the constituents of the beer, and on the pH, being a minimum for the normal pH range of beer. Iron and copper, and probably also phosphate, calcium, ammonium and hydroxyl ions and yeast co-enzyme, are active oxygen-carriers in these oxidation processes; and it is believed that organic compounds containing the grouping $-C(OH)=C(OH)-$ are also involved in autoxidation. Indicator—time tests for beer stability suggest that the stability decreases if the wort is oxidised before fermentation (e.g. as in the hot aeration process), and that storage at a high temperature or slow cooling after pasteurisation, accelerates oxidation; agitation also increases haze-formation. The stability is increased as the result of the boiling operation, and the simultaneous increase in colour suggests that the melanoidins may contribute to the reducing properties, which, however, are little influenced by the fermentation process. Beer which contains 5 to 7 ml. of air per bottle will not keep for more than half as long as beer in air-free bottles. J. G.

Determination of Iron in Bread and Bread Ingredients. C. Hoffman, T. R. Schweitzer and G. Dalby (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 454-455.)—Loss of iron during ashing is entirely prevented by addition of 5 ml. of N sodium hydroxide solution per 1 g. of sample. The weighed substance is thoroughly mixed with the lye, dried at 100° C., and ashed at a low red heat in an electric muffle. The iron in the ash is determined colorimetrically as usual. W. R. S.

Body Fats of the Pig. V. Component Glycerides of Perinephritic and Outer Back Fats from the same Animal. T. P. Hilditch and W. H. Pedelty. (*Biochem. J.*, 1940, 34, 971-979.)—The pig fats were examined by a method similar to that previously used (Hilditch and Paul, *Biochem. J.*, 1938, 32, 1775) in the examination of ox depot fat. The fats were systematically crystallised from acetone and grouped into three or four fractions according to their iodine values, and the respective component acids were determined by ester fractionation. The amounts of fully-

saturated glycerides in the two fractions most sparingly soluble in acetone were then estimated by oxidation with potassium permanganate in acetone solution, and, when possible, their component acids were estimated by ester fractionation. The fractions most soluble in acetone (in which the di- and tri-unsaturated glycerides are concentrated) were completely hydrogenated, and the products were fractionated with ether. The component acids in each sub-fraction so obtained were estimated by ester-fractionation, and the component glycerides in each were calculated as binary mixtures of tristearin and palmitodistearin, or of palmitodistearin and dipalmitostearin, according to the proportions of palmitic and stearic acid found. From these results, the amount of tristearin in the hydrogenated product was calculated, and thence the amount of tri- C_{18} glycerides in the acetone-soluble fractions of the original fat. The data thus obtained permit the component glycerides of each fraction of the fat to be grouped into (a) dipalmitomono- C_{18} , palmitodi- C_{18} and tri- C_{18} glycerides, and (b) fully-saturated, mono-unsaturated-disaturated, di-unsaturated-mono-saturated and tri-unsaturated glycerides. From the proportions of these classes of glycerides, the individual components in each fraction of the original fat were deduced. The minor fatty acids present were included with the appropriate major acid. The method now introduced is believed to be much more accurate than those used in the past, and it is claimed that a practically complete statement of the component glycerides in animal depot fats can be given with a fair chance of certainty. The molar percentages of the main components of the two pig fats were found to be:

| | Outer back fat | Perine- phritic fat |
|----------------------------|----------------------|---------------------------|
| Palmitodi-"olein" .. | 46-53 | 35-40 |
| "Oleo"-palmitostearin .. | 34-27 | 39-34 |
| "Oleo"-dipalmitin .. | 5 | 9 |
| Stearodi-"olein" .. | 0-7 | 0-5 |
| Tri-"oleins" .. | 10-3 | 8-3 |
| Fully saturated glycerides | 5 | 9 |

The results now obtained, together with those on ox depot fat previously reported, support the hypothesis that stearo-glycerides are formed in the animal body by the saturation of pre-formed oleo-glycerides. In addition, a gradual increase in the proportions of dipalmito-glycerides is observed as the general degree of saturation increases.

F. A. R.

Detection of Saccharin in Foodstuffs. W. F. Reindollar. (*J. Assoc. Off. Agr. Chem.*, 1940, 23, 288-290.)—The two tests for saccharin given in the *Methods of Analysis* of the A.O.A.C. depend upon the sweet taste of

the ethereal extract and conversion of the saccharin into salicylic acid. Neither test is specific, since other extracted matter may have a sweet taste, and the procedure in the second test does not destroy naturally occurring or added salicylic acid. The following test, depending upon the conversion of saccharin into phenolsulphonaphthalein, is recommended: The sample (25 g.) is mixed with sufficient boiling water to produce 75 ml. and, after standing for 1 hour with occasional shaking, the mixture is acidified with 3 ml. of glacial acetic acid, and 5 ml. of 20 per cent. neutral lead acetate solution are added. The mixture is diluted to 100 ml., allowed to stand for 20 minutes and filtered, and an aliquot portion (60 ml.) of the filtrate is acidified with 5 ml. of hydrochloric acid and extracted with a mixture of equal parts of ether and petroleum spirit (b.p. 30° to 60° C.). The ethereal solution is washed with 5 ml. of water and, after preliminary concentration, the solvent is allowed to evaporate at room temperature. The residue is heated for 2 hours at 135° to 140° C. with 5 ml. of a solution of crystalline phenol in an equal weight of sulphuric acid, and the product is dissolved in a little hot water and poured into about 250 ml. of cold water. After 3 hours the liquid is filtered, made alkaline with 10 per cent. sodium hydroxide solution, and diluted to 500 ml. A magenta or reddish purple colour indicates the presence of saccharin in the original sample. A yellow, buff, or pale salmon colour is not significant, and will be given by a blank experiment made with similar material. Vanillin, if present, must be removed by repeated extraction of the ethereal extract with carbon tetrachloride. With some foods (e.g. ginger ale) the preliminary treatment with lead acetate is unnecessary. The method gives satisfactory results with foodstuffs containing 80 p.p.m. of saccharin (equivalent in sweetening power to 4 per cent. of sugar), and will probably detect smaller amounts. A. O. J.

Determination of Vanillin in Vanilla Extracts. H. J. Lynch and N. Deahl. (*J. Assoc. Off. Agr. Chem.*, 1940, 23, 429-431.)—It was found that in the determination of vanillin in concentrated vanilla extracts containing added vanillin by the official colorimetric method (*Methods of Analysis* of the A.O.A.C., 1935, 307) a considerable portion of the vanillin was not recovered, and the method was examined to discover the cause of its failure. In a modification of the official method the extract containing 8 to 12 mg. of vanillin (usually about 5 ml.) was diluted with water so that a further addition of 4 ml. of the solution of neutral and basic lead acetates produced 50 ml. The mixture was filtered and the precipitate was washed with water until the filtrate measured 100 ml. A 5-ml. portion of this filtrate was assayed by the official method, but, although higher results were obtained, the whole of the vanillin was not found. Subsequent investigation showed that the vanillin could be removed quantitatively

from the lead precipitate only by the use of an inconveniently large amount of washing water, and the results indicated that precipitation should be made in a much more dilute solution. The following method is proposed: The sample, containing 8 to 12 mg. of vanillin, is diluted to 100 ml. with water, and a 5-ml. portion of the dilute solution is treated with 0.2 ml. of a solution containing 50 g. each of the neutral and basic acetates of lead per litre. A standard vanillin solution (5 ml.) containing 0.1 mg. of vanillin per ml. is used as control. To each of the solutions 5 ml. of the vanillin reagent (Folin and Denis, *J. Ind. Eng. Chem.*, 1912, 4, 670; *Abst., ANALYST*, 1912, 37, 501) are added, and after 5 minutes the mixtures are diluted to 50 ml. with saturated sodium carbonate solution. Finally the liquids are allowed to stand for 10 minutes and filtered, and the blue colours are compared in a colorimeter. By this method 99.5 per cent. of the known vanillin-content of a sample was found, compared with 79.46 per cent. by the official method. It would appear that in the official method originally developed by Folin and Denis (*loc. cit.*) vanillin is lost either by precipitation as a lead compound or by adsorption on the lead resinates. It is generally accepted that a good vanilla extract should contain about 0.2 g. of vanillin per 100 ml. when assayed by the official method. Such extracts yield over 0.3 per cent. of vanillin by the modified method. A. O. J.

Biochemical

Estimation of Manganese in Organic Material containing Large Amounts of Calcium and Chlorides. T. W. Ray. (*J. Biol. Chem.*, 1940, 134, 877-881.)—Studies on the metabolism of manganese in the mouse necessitated the estimation of manganese in the ash of the whole animal. The dead mice were dried in an electric oven at 105° C. and ashed in silica beakers in a silica-lined muffle-furnace maintained thermostatically at a temperature not exceeding 700° C. After 8 hours' heating, the ash was allowed to cool and dissolved in 10 ml. of conc. nitric acid. Four drops of conc. sulphuric acid were added (not more, to avoid the formation of more calcium sulphate than could be dissolved), and the solution was evaporated to dryness over a free flame to remove the chlorides. The residue was dissolved in 12 ml. of 25 per cent. nitric acid, and the solution was diluted to 20 ml. (or more, if the mice had been fed on a manganese-rich diet). To an aliquot portion of the solution were added 0.2 ml. of 0.25 per cent. silver nitrate solution and 0.25 g. of potassium persulphate. The mixture was then heated in a water-bath for a few minutes to develop the full permanganic acid colour; it was compared with a standard similarly treated. The most accurate readings were obtained with not more than 0.02 mg. of manganese in 10 ml. of solution. The recovery of added manganese varied between

96 and 103 per cent. of the theoretical. The method was also applied successfully to the estimation of manganese in milk. F. A. R.

Determination of Fumarate and Malate in Animal Tissues. H. A. Krebs, D. H. Smyth and E. A. Evans. (*Biochem. J.*, 1940, **34**, 1041-1045).—The polarimetric method for the determination of *l*(-)-malate is not specific. The following method, based on reduction of fumarate to succinate, which is then estimated manometrically with succinic dehydrogenase, can be applied to quantities of fumarate from 0.05 mg. upwards; the concentration of malate is calculated from the equilibrium constant. The tissue suspension is deproteinised at 40° C., by adding one-fifth of its volume of 5 per cent. metaphosphoric acid, and filtered. An aliquot portion of the filtrate is transferred to a measuring cylinder, and 0.5 g. of zinc filings, 2.3 ml. of 10 *M* phosphoric acid and 0.25 ml. of copper sulphate solution (20 per cent. solution of $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$) are added to each 10 ml. of solution. After one hour the succinic acid is extracted with ether in a continuous extractor. The extract is treated with 1 ml. of 0.1 *M* disodium phosphate solution, and the ether is removed by distillation. The residue is transferred to a measuring cylinder, adjusted to pH 7 and made up to a known volume (1 to 3 ml.). The succinate in the solution is estimated manometrically with succinic dehydrogenase (Krebs, *Biochem. J.*, 1937, **31**, 2095). The concentration of *l*(-)-malate is calculated from the equilibrium constant. At pH 7.4 the ratio *l*(-)-malate/fumarate was found to be 2.65 at 50° C., 3.17 at 40° C., 3.54 at 30° C., and 4.57 at 20° C. Malate, tartrate, oxaloacetate, glutamate, citrate and aconitate do not yield succinate under these conditions, but malate (not present in biological material) behaves like fumarate. F. A. R.

Estimation of Nicotinic Acid in Biological Materials by means of Photoelectric Colorimetry. D. Melnick and H. Field. (*J. Biol. Chem.*, 1940, **134**, 1-16).—Several workers have used the colour produced by treatment with cyanogen bromide and an aromatic amine for estimating nicotinic acid in foodstuffs (*cf.* ANALYST, 1939, **64**, 441, 755; 1940, **65**, 182, 183), and this method has been adopted in the present instance. The preliminary treatment has been modified, however, direct acid hydrolysis of the test substance being used, followed by preferential charcoal adsorption for the decolorisation of the hydrolysate. The test material, containing 10 to 400 γ of nicotinic acid, is treated in a test-tube, graduated at the 10- and 15-ml. marks, with 5 ml. of conc. hydrochloric acid and water to make the volume up to 15 ml. The test-tube is immersed in boiling water for 30 to 40 minutes, then cooled, and the volume is adjusted to the original 15 ml. Ten ml. of absolute ethyl alcohol are added, and the solution is transferred to a 150-ml. conical flask. Exactly 200 mg. of adsorbent charcoal

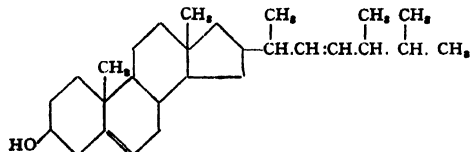
are added, and the mixture is shaken and filtered at room temperature. An aliquot part of the filtrate (8.33 ml.) is pipetted into the graduated test-tube, a drop of phenolphthalein solution is added, and the solution is neutralised in the cold to pH 7 with conc. sodium hydroxide solution, and its volume is adjusted to 10 ml. A 3-ml. aliquot portion is mixed with 7 ml. of an alcoholic buffer solution of pH 2.4 (1960 ml. of water, 10 ml. of 85 per cent. phosphoric acid, 30 ml. of 15 per cent. sodium hydroxide solution and 333 ml. of absolute ethyl alcohol), and the colour of the solution is measured in an Evelyn photoelectric colorimeter, filter 420 being used. A second 3-ml. aliquot portion is treated with 6 ml. of cyanogen bromide reagent (water saturated with bromine at 5 to 10° C. is just decolorised in the cold by means of 10 per cent. potassium cyanide solution) from a burette, followed immediately by 1 ml. of a 4 per cent. solution of aniline in absolute ethyl alcohol. After mixing, the colour of the solution is measured in the colorimeter with the same filter as before. The reading given by the extract alone is subtracted from that obtained after addition of the reagents, and the difference gives the photometric density produced by the nicotinic acid and nicotinamide present. The amount of nicotinic acid cannot, however, be calculated from a standard curve, owing to the presence of different amounts of inhibitors in the extract. A third 3-ml. aliquot portion of the extract is therefore treated with 10 γ of nicotinic acid, and the procedure is repeated. The difference between this and the second reading gives the photometric density produced by 10 γ of nicotinic acid, and from this value the amount of nicotinic acid present in the extract is calculated. In contrast to those described in previous reports, the reagents thus prepared were stable for at least 5 months. The recovery of known amounts of nicotinamide added to biological materials varied from 97 to 108 per cent. in 14 experiments and was usually between 99 and 102 per cent. F. A. R.

Coupled Oxidation of Carotene and Fat by Carotene Oxidase. J. B. Sumner and R. J. Sumner. (*J. Biol. Chem.*, 1940, **134**, 531-533).—The name "carotene oxidase" was given to an enzyme which peroxidises unsaturated fats and bleaches carotene, bixin, xanthophyll and other carotenoids; it is present in legumes, especially in soya beans. In earlier experiments the carotene was dissolved in an unsaturated oil, and under these conditions bleaching was rapid. When, however, a solution of carotene in a mixture of acetone and alcohol was added to the enzyme extract, bleaching of the resulting carotene suspension was very slow; thus 0.79 mg. of carotene required 18 to 20 hours without oil, and only 32 seconds when mixed with 4.6 mg. of hempseed oil. The presence of additional fat reduced the rate of bleaching. Unsaturated fats were much more effective, weight for weight, than saturated fats. Fat previously peroxidised by carotene oxidase had no

increased action on the bleaching of carotene, nor did it cause immediate bleaching when carotene was dissolved in it; to exert its effect the fat must be mixed with the carotene before being suspended in water. Thus the bleaching action of carotene oxidase appears to be a coupled reaction.

F. A. R.

Brassicasterol, the Characteristic Sterol of Rapeseed Oil. E. Fernholz and H. E. Stavely. (*J. Amer. Chem. Soc.*, 1940, **62**, 1875-1877.)—Brassicasterol, m.p. 148° C., was first isolated by Windaus and Welsch from the sterol fraction of rapeseed oil *via* the sparingly soluble acetate tetrabromide. This was difficult to distinguish from the acetate tetrabromide of stigmasterol, but on debromination it yielded brassicasterol acetate, m.p. 158° C. (stigmasterol acetate melts at 142° C.). The constitution of brassicasterol has now been elucidated. Ozonolysis of a carefully purified specimen yielded methyl isopropyl acetaldehyde, identical with the aldehyde formed under similar circumstances from ergosterol. On catalytic hydrogenation, ergosterol and brassicasterol give the same saturated sterol, ergosterol. Thus brassicasterol is 7:8-dihydro-ergosterol with the formula:



F. A. R.

Ascorbic Acid Oxidase from Summer Crook-neck Squash (*C. pepo condensae*). P. L. Lovett-Janison and J. M. Nelson. (*J. Amer. Chem. Soc.*, 1940, **62**, 1409-1412.)—The existence of the so-called ascorbic acid oxidase has been doubted by some workers, who attribute the oxidation of ascorbic acid to the catalytic effect of traces of copper or a copper-protein complex. Evidence is now put forward suggesting that ascorbic acid oxidase is a true enzyme, consisting of a copper-protein, similar in type to tyrosinase. After several possible sources had been examined, summer crook-neck squash was chosen as the best source of the active substance. The juice, obtained by expression of the minced rinds, was freed from inert material by treatment with barium acetate solution, and the filtrate, freed from excess of barium, was treated with ammonium sulphate until 0.6 saturated. The precipitate, which was active, was treated with disodium hydrogen phosphate solution, and the filtrate was made 0.9 saturated with magnesium sulphate. This treatment precipitated about two-thirds of the enzyme, and the remainder was precipitated by fully saturating the filtrate with magnesium sulphate. It was not possible to effect further concentration of the first precipitate, but the second precipitate was treated as follows:—It was dialysed and adsorbed on alumina, from which it was eluted with secondary sodium phosphate solution.

The eluate was dialysed and treated with 0.1 saturated lead acetate solution and acetone cooled in solid carbon dioxide. The precipitate formed was removed in the centrifuge and the supernatant liquid was re-treated, the procedure being repeated three times in all. The combined precipitates were dissolved in secondary sodium phosphate solution and the solution was subjected to further adsorption, elution, dialysis and fractionation until no further concentration resulted. The copper content of the enzyme was estimated by the method of Warburg (*Biochem. Z.*, 1927, **187**, 255) at various stages in the purification. A mixture of 2 ml. of 0.2M sodium pyrophosphate solution (pH 7.75), 0.1 ml. of 0.2N hydrochloric acid and 0.2 ml. of a solution containing 6 mg. of cysteine hydrochloride was introduced into each of the side-arms of six Warburg respirometers. To the first was added 0.1 ml. of a solution containing 0.1γ of copper, to the second 0.2 ml. of a similar solution, and to the fourth, fifth and sixth, 0.1, 0.2 and 0.3 ml. respectively of the enzyme solution. The third was a blank with reagents but no enzyme. Each mixture was then diluted to 2.6 ml. with water. When the temperature of the bath was constant at 25° C., the contents of each side-arm were transferred to the flasks of the apparatus containing a buffer solution of pH 7.6. Manometer readings, taken at 5-minute intervals, were converted into c.mm. of oxygen. By subtracting the value for flask 4 from those for flasks 1 and 2, the uptakes due to 0.1 and 0.2γ of copper were obtained; and by subtracting the value for flask 4 from those for flasks 5 and 6, the uptakes due to 0.1 and 0.2 ml. of enzyme were obtained. The uptake due to 0.1 ml. of enzyme solution divided by the uptake due to 0.1 ml. of copper solution gave the concentration of copper in the enzyme solution. It was found that the percentage of copper in the preparation increased as the enzyme activity increased until it reached a value of 0.15 per cent. This value was not exceeded in spite of further attempts at purification. The correspondence between copper content and enzyme activity is strong evidence for the conclusion that the active group of ascorbic acid oxidase contains copper. The purified enzyme has 1100 times the activity of copper alone, 13,000 times the activity of a copper-albumin mixture and 4100 times the activity of a copper-gelatin mixture, suggesting that the purified material is, indeed, a true enzyme. In view of Theorell's observation that dihydroxymaleic acid oxidase contains manganese as activator, the presence of manganese was sought in the ascorbic acid oxidase, but the amount was found to decrease as the enzyme activity increased. Similarly, the final product was almost completely free from peroxidase activity. The enzyme had little or no action toward *p*-cresol, catechol or hydroquinone.

F. A. R.

Effect of Zinc on Alkaline Phosphatases. E. Hove, G. A. Elvehjem and E. B. Hart. (*J. Biol. Chem.*, 1940, **134**, 425-442.)—Bone

phosphatase preparations were made by grinding cleaned fresh bone (rat femur) and allowing the suspension to autolyse for 24 hours; the filtered solution contained about 4 mg. of dry matter per ml. Preparations of intestinal phosphatase were similarly prepared from minced rat intestine; after dialysis the solution contained 2 mg. of dry matter per ml. The activity of the phosphatase preparations was measured with sodium β -glycerophosphate as substrate, but disodium phenyl phosphate, sodium hexose diphosphate and sodium pyrophosphate were also used. The phosphate esters were made up as a $4.8mM$ solution (1 ml. = 0.15 mg. of phosphorus) in a $50mM$ veronal buffer solution, the final pH being adjusted to 9.2. Ten ml. of the substrate solution and 0.5 ml. of enzyme solution were made up to a total volume of 11.5 ml. and incubated at $37^{\circ}C$. The reaction was stopped by adding 2 ml. of 10 per cent. trichloroacetic acid solution, and the inorganic phosphorus produced in the reaction was estimated by the method of Fiske and Subbarow (ANALYST, 1926, 51, 205). It was found that crude intestinal phosphatase activity was increased 40 to 100 per cent. by addition of zinc ions, whereas crude kidney and bone phosphatase activities were progressively inhibited by concentrations of zinc of 4×10^{-3} to $70 \times 10^{-3}mM$; this effect was independent of the nature of the substrate. The effect of zinc and magnesium together was greater than that produced by either alone. Dialysis of the crude intestinal phosphatase resulted in a preparation that was not activated by zinc ions, but which, like the other two phosphatase preparations, showed progressive inhibition. The dialysate, when added to the dialysed intestinal enzyme, restored the original antivitality with zinc, but the dialysate alone had no effect on the substrate. The zinc co-activator present in the dialysate was a product of mucosal tissue autolysis, and its effect was simulated by a casein hydrolysate. Subsequently it was shown that all α -amino acids behave as zinc co-activators, though β -amino-, keto- and hydroxy-acids and aliphatic acids or organic amines have little or no effect. The optimal concentration of the amino-acids was studied in detail for glycine; it was found that the optimal zinc and optimal glycine concentrations were interdependent, optimal stimulation being obtained with 1 mole of zinc for every 75 to 100 moles of glycine. At high glycine concentrations the zinc optimum was broad and flat, whereas at lower glycine concentrations it was sharp. The phosphatases of the bone and intestines of zinc-deficient rats were compared with similar preparations from normal animals. Whereas the activities of the bone phosphatases differed little, the intestinal phosphatase activity of zinc-deficient rats was considerably lower than that of normal controls. These results probably explain why zinc is essential to the life and growth of rats; the lowered blood non-protein nitrogen in zinc deficiency would also be explained. Moreover, if it is true that amino-acid absorp-

tion involves phosphorylation, the slower amino-acid absorption observed with zinc-deficient rats would be explained by the lowered intestinal phosphatase activity of these rats. F. A. R.

Identification of the Rice Factor. H. J. Almquist, E. L. R. Stokstad, E. Mecchi and P. D. V. Manning. (*J. Biol. Chem.*, 1940, 134, 213-216.)—Chicks reared on a diet consisting of casein, water-washed sardine-meal, yeast, wheat-germ oil or soya bean oil, minerals, vitamins A and D, glucose or starch and sucrose grew more rapidly when the pure carbohydrates were replaced by polished rice, or when cartilage was added to the basal diet. It was subsequently found that gelatin or synthetic glycine in combination with chondroitin resulted in a rate of growth equal to that produced by the cartilage supplement. Glycine or chondroitin, given separately, did not effect the same growth increase as the two substances administered together. Thus the "rice factor" is identical with, or at least is replaceable by, a mixture of glycine and chondroitin. F. A. R.

Oxidation of Vitamin E. C. Golumbic and H. A. Mattill. (*J. Biol. Chem.*, 1940, 134, 535-541.)—It has been suggested that the irreversible oxidation of a phenolic substance may occur in two steps, the initial step being reversible and consisting in the production of a phenoxyl radical; this forms an unstable oxidation-reduction system with the phenol. When such a reversible step governs the rate of the irreversible oxidation of a phenol, it is possible to measure an apparent oxidation potential by potentiometric indicators. Such an apparent oxidation potential (the potential at which 20 to 30 per cent. was oxidised in 30 minutes) of α -tocopherol was measured by two different methods and was found to lie between the normal oxidation potentials of mono- and dimethylhydroquinones. α -Tocopheryl quinone, the oxidation product of α -tocopherol, was found to be biologically inactive when pure. Previous statements to the contrary are explained by the presence of unchanged α -tocopherol due to incomplete oxidation; with gold chloride, but not with ferric chloride, oxidation is complete at room temperature. Pure α -tocopheryl quinone is also without antioxygenic action on lard. Various substituted hydroquinones with oxidation potentials of the same order as α -tocopherol were found to be biologically inactive, alone or with phytol. These observations indicate that the reaction: α -tocopherol \rightarrow α -tocopheryl quinone is irreversible in the organism, but it may take place in two steps, of which the first is reversible. This may be of biological importance. F. A. R.

Vitamin-free Diets for Animal Experiments. A. L. Bacharach. (*Nature*, 1940, 146, 28-29.)—The difficulty of obtaining supplies of rice starch for use as the sole or predominant source of carbohydrate for basal

diets required in work on vitamin A, vitamin B, or vitamin E may be overcome by the use of wheat starch. Experiments illustrating this conclusion are described. Wheat starch cannot, however, satisfactorily replace rice starch for diets used in work on the rat "filtrate factor," discrimination between negative controls and animals receiving filtrate factor concentrate made from fresh liver being seldom satisfactory. It is believed that the wheat starch used had adsorbed or otherwise retained appreciable quantities of filtrate factor from the wheat berry. No corresponding information is available concerning the effect of wheat starch on diets used for work on vitamin B₆ (adermin, "eluate factor"). J. G.

Bacteriological

Molecular Constitutions of Catenarin and Erythroglauclin. W. K. Anslow and H. Ralstrick. (*Biochem. J.*, 1940, **34**, 1124-1133).—Catenarin, C₁₈H₁₆O₆, which constitutes 15 per cent. of the dry weight of the mycelium of *Helminthosporium catenarium* Drechsler, is 1 : 4 : 5 : 7 - tetra - hydroxy - 2 - methyl-anthraquinone. Erythroglauclin, C₁₈H₁₄O₆, which is a metabolic product of species in the *Aspergillus glaucus* series, is the 7-methyl ether of catenarin, i.e. 1 : 4 : 5-trihydroxy-7-methoxy-2-methylanthraquinone. F. A. R.

Agricultural

Plant Insecticide Materials from Empire Sources. (*Bull. Imp. Inst.*, 1940, **38**, 150).—Derris: Wide variations in the composition of *D. elliptica* and *D. malaccensis* roots from different countries have been found as follows:

| No. of samples | Country of origin | Moisture | Total extract (etheral) | Rotenone calculated from carbon tetrachloride samples | Rotenone purified by alcohol | |
|----------------|-------------------|--------------|-------------------------|---|------------------------------|------|
| | | | | | Range | Mean |
| 8 | Tanganyika | 4.5 to 10.2 | 3.3 to 22.6 | 1.0 to 11.0 | 0.95 to 9.3 | 6.3 |
| 3 | Sarawak .. | 10.0 .. 10.6 | 10.3 .. 13.1 | 1.7 .. 3.6 | 1.3 .. 3.3 | 2.3 |
| 6 | Seychelles .. | 5.9 .. 7.9 | 8.1 .. 18.4 | 1.2 .. 7.0 | 1.0 .. 6.5 | 3.4* |
| 1 | " .. | 6.1 | 14.9 | 3.1 | 2.6 | |
| 5 | Trinidad .. | 4.9 .. 7.6 | 9.0 .. 22.0 | 1.1 .. 8.6 | 0.8 .. 7.2 | 3.8 |
| 3 | Dominica .. | 6.0 .. 8.3 | 3.8 .. 13.3 | 0.8 .. 4.6 | 0.6 .. 3.6 | 2.3 |
| 2 | Fiji .. | 9.8 .. 9.9 | 6.9 .. 9.3 | 1.5 .. 1.8 | 1.3 .. 1.6 | 1.45 |
| | | | (Chloroform) | | | |
| 1 | Tanganyika | 7.2 | 18.0 | — | 7.6* | |
| 1 | Mauritius .. | 7.1 | 12.6 | — | 4.85* | |

* Purified by trituration with cold alcohol (other samples crystallised from hot).

Mundulea sericea (or *M. suberosa*).—Two South African Union samples from root and bark respectively contained no rotenone or only traces. Owing to the reported toxicity of this plant, which was formerly attributed to rotenone, biological tests were made at Rothamsted with *Aphis rumicis*. An alcoholic root extract was completely toxic at 1 per cent. concentration (in terms of root used). Other results were:—Concentrations (as percentages) 0.5, 0.25, 0.1: moribund and dead insects (per

cent.), 50, 20, 10. Stems and bark of the same plant gave negative biological tests, but stems from an Indian sample were toxic, and *M. suberosa* leaves from India were completely toxic at 1 per cent. One sample of Tanganyika bark, rotenone-free, contained saponins and alkaloids; a second contained 0.7 per cent. of rotenone (0.8 per cent. on moisture-free material) and ethereal extract, 4.6 per cent.

Tephrosia vogelii (Uganda): Leaves from a Toro estate (1a), from white-flowered Kampala Plantation plants (1b) and from purple-flowered Kampala Plantation plants (1c) gave alcoholic extracts which, on dilution with aqueous solutions of saponin (0.5 per cent.) to 5 per cent. alcoholic solutions, were toxic to *Aphis rumicis* as follows:—Concentration of leaf, g./100 ml.: 1.0, 0.5, 0.25, 0.1, 0.05; percentage of insects paralysed: (1a) 100, 100, 25, 5, 5; (1b) 100, 100, 25, 10, 0; (1c) 100, 100, 85, 5, 5 (control spray toxicity 3 per cent.). *Tephrosia toxicaria* (Natal) roots contained 1 per cent. of ethereal extract and only a trace of rotenone. Insecticidal properties are due to tephrosin.

Pyrethrum: Samples of flowers harvested at different stages, containing various amounts of stalk, and obtained from different countries, were analysed. Two of the St. Helena samples—12a, dried flowers; 12b, dried stalks—were grown experimentally from seed supplied by Kew Gardens in 1934 and examined in 1937. Pyrethrin I (0.46 per cent.) and pyrethrin II (0.56 per cent.) of the flowers were satisfactory (moisture, 8.6 per cent.); the stalks contained only 0.05 per cent. of each. Other results (as percentages) were: *Tanganyika* (14 samples): moisture, 7.0 to 10.7; pyrethrin I, 0.38 to 0.64; pyrethrin II, 0.33 to 0.80; total

pyrethrins, 0.73 to 1.40. A sample of pyrethrum powder contained: moisture, 10.7; pyrethrin I, 0.30; pyrethrin II, 0.32; total pyrethrins, 0.62 per cent. *St. Helena* (1 sample): moisture, 10.0; pyrethrin I, 0.45; pyrethrin II, 0.59; total pyrethrins, 1.04 per cent. *Ceylon* (1 sample): moisture, 7.3; pyrethrin I, 0.47; pyrethrin II, 0.57; total pyrethrins, 1.04. There is an optimum time for harvesting.

Chrysanthemum frutescens (Tanganyika, 1

sample): moisture, 9.1; pyrethrin I, 0.05; pyrethrin II, 0.07; total pyrethrins, 0.12 per cent. Biological tests on this were negative. In appearance the plant resembled normal pyrethrum. E. B. D.

Toxicity to Sheep of Lead Arsenate and Lead Arsenate Spray Residues. J. L. St. John, E. C. McCulloch, J. Sotola and E. N. Todhunter. (*J. Agr. Res.*, 1940, 60, 317-329.)—An investigation of outbreaks of illness or death of cattle and sheep fed in autumn on pasture in sprayed orchards showed that they were due to chemical poisoning; approximately 15 per cent. of the poisoned animals recovered. Orchard grasses from an area where approximately 1000 sheep died contained 0.44 per cent. of arsenic and 1.44 per cent. of lead. Experiments with healthy lambs (average wt., 80 lbs., or 36.29 kg.) showed that the total lethal dose of arsenic, as lead arsenate, was 1.5 g. (about 41.0 mg. per kg.) when given daily in small amounts (0.25 to 2 g.) in gelatin capsules. Lead was determined by the 1935 tentative methods of the Association of Official Agricultural Chemists, the Gerritz preliminary treatment being applied (*cf. Ind. Eng. Chem., Anal. Ed.*, 1935, 7, 167) and colorimetric comparisons being made. Arsenic was determined by the bromate or the Gutzeit method. The lead and arsenic recovered from the total urine and faeces, from the stomach contents, and from a number of organs amounted to less than the quantity consumed. Maximum recovery was from the stomach contents, urine and faeces, the proportion of lead eliminated being larger than that of arsenic, elimination of arsenic per day being greater in urine than in faeces with larger consumption and *vice versa* with smaller amounts. Lead, which was eliminated mainly in the faeces, was always proportionally higher than the arsenic in these. The experimental animals were fed with a ration of rolled oats, rolled barley and alfalfa hay, in proportions 3 : 4 : 14 (salt *ad lib.*). Addition of apples to the ration apparently did not alter toxicity of lead and arsenic. Lead arsenate on sprayed foliage became less toxic after several weeks than the experimental capsules, and investigation of possible change in form of the lead and arsenic is required. E. B. D.

Gas Analysis

Elimination of Traces of Carbon Monoxide from Hydrogen. K. Kawakita and B. Ichimiyagi. (*J. Soc. Chem. Ind. Japan*, 1940, 43, 121B.)—The gas is passed at ordinary pressure over activated reduced iron at 350° to 400° C., the carbon monoxide being decomposed into carbon and carbon dioxide, which latter is chemically absorbed. For the detection of traces of carbon monoxide the hydrogen was passed through blood solutions, the absorption spectrum of which was examined before and after treatment. The quantitative determination of the carbon monoxide was carried out by oxidation with iodine pentoxide

(temperature not given), which gave satisfactory results with carbon monoxide contents lower than 0.04 per cent. With a catalyst obtained by reduction of 8 g. of ferric oxide, the carbon monoxide was reduced to about 0.001 per cent. at 360° C. at a velocity of 4 litres per hour; it could no longer be detected with a catalyst obtained from 16 g. of ferric oxide and a gas velocity of 5 litres per hour at 360° C. W. R. S.

Organic

Quantitative Determination of Certain Polyalcohols in Presence of Each Other. N. Allen, H. Y. Charbonnier and R. M. Coleman. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 384-387.)—The reaction of periodic acid with polyalcohols (*cf. Malaprade, ANALYST*, 1928, 53, 299) has been applied to the determination of, and to distinguish between, certain polyalcohols, notably glycerol and ethylene glycol. These two polyalcohols may also be determined in presence of one another by an acidimetric-iodimetric titration, or potentiometrically with the glass electrode, and a combination of the Malaprade reaction and an acid dichromate oxidation enables solutions containing three glycols to be analysed. A quantity of sample containing oxidisable matter corresponding with the equivalent of not more than 0.09 g. of glycerol is diluted to 50 ml. and allowed to stand for 40 to 80 minutes in a glass-stoppered flask at 20° C. with 50 ml. of a filtered 0.05 N solution of periodic acid (calculated as a monobasic acid, *i.e.* 11 g. per litre). When oxidation is complete, 100 ml. of water are added, followed by 20 ml. of 0.1 N sodium hydroxide solution, 2 drops (not more) of methyl red indicator, and then sufficient of the 0.1 N alkali to destroy the pink colour. To the titrated solution are added 150 ml. of water, 30 ml. of 20 per cent. potassium iodide solution and 25 ml. of 6 N sulphuric acid, and the mixture is titrated with 0.2 N sodium thiosulphate solution with starch as indicator; the end-point is sharp, but the final solution has a pink shade owing to the methyl red. Allowance is made for the blank obtained with 50 ml. of water, and since 1 ml. of N sodium hydroxide \equiv 0.09206 g. of glycerol (acidimetric titration), the glycerol content (*G* per cent.) may be calculated. Since, further, 1 ml. of N sodium thiosulphate solution \equiv 0.023015 g. of glycerol (iodimetric titration), this titration enables the joint percentage of glycerol and ethylene glycol (*T*) to be determined in terms of glycerol; then the ethylene glycol content is 1.348 (*T-G*) per cent. If a third glycol, which is not oxidised by periodic acid (*e.g.* diethylene glycol) is present, it is determined by oxidation with dichromate. Thus the sample (half the quantity previously taken) is diluted to 25 ml. and boiled under a reflux condenser for 20 minutes with 25 ml. of a 2.4 per cent. solution of potassium dichromate and 40 ml. of conc. sulphuric acid. The cooled solution is then diluted to 300 ml., 10 ml. of 20 per cent.

potassium iodide solution are added, and the iodine is titrated with 0.2 *N* sodium thiosulphate solution in the usual way, allowance being made for a blank. Then, since 1 ml. of *N* sodium thiosulphate solution = 0.006576 g. of glycerol, the combined glycerol, ethylene glycol, and diethylene glycol contents may be calculated in terms of glycerol (*X* per cent.); then the diethylene glycol-content is 0.807 (*X-T*) per cent. For the potentiometric method 50 ml. of the 0.05 *N* periodic acid solution are diluted with 150 ml. of water and titrated with 0.1 *N* sodium hydroxide solution; the first equivalent-point (at about pH 5.5) is determined (*B* ml.), an equal volume of reagent is added, and the resulting pH (about 10.0) is determined. The sample (*w* g.) is then oxidised with the periodic acid as described above, and after dilution with 100 ml. of water the solution is titrated with the 0.1 *N* alkali to the first equivalent-point (*A* ml.). Titration is then continued to the pH value found for the second equivalent-point of the blank (*X* ml.). Then $0.9206(A-B)/w$ = percentage of glycerol; and $0.6205(3B-X-A)/w$ = percentage of ethylene glycol. Results tabulated for a number of mixtures of the above polyalcohols in known proportions record errors ranging from -4.68 to +12.94 per cent.; the error for the determination of the ethylene glycol is always greater than for glycerol, since the former is not determined directly. The acidimetric method for glycerol may be used in presence of a variety of organic compounds, so long as these do not react with periodic acid to form an acid. Unknown solutions of polyalcohols should first be oxidised with periodic acid, and the results of the acidimetric iodimetric titrations calculated in terms of glycerol. If these figures agree with that obtained from the dichromate oxidation and calculated in the same way, glycerol is the only polyalcohol present; if the values are different a mixture of polyalcohols is indicated. The fact that glycerol reacts with periodic acid to produce formic acid, whereas ethylene glycol does not, is the basis of the following qualitative test to distinguish between them when they are the only two constituents present. The solution (2 ml.), containing a drop of methyl red, is adjusted with acid or alkali until it is faintly acid to the indicator; 2 ml. of a solution of periodic acid are treated similarly. If a pink colour develops when the solutions are mixed, glycerol is present. J. G.

Determination of Unsaturation in Aliphatic Hydrocarbon Mixtures by Bromine Absorption. J. B. Lewis and R. B. Bradstreet. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 387-390.)—The Francis method (*ANALYST*, 1926, 51, 534) has been modified as follows (see also Thomas, Block and Hoekstra, *Ind. Eng. Chem., Anal. Ed.*, 1938, 10, 153):—From 0.7 to 1.0 g. of the sample is added from a Lunge weighing-pipette to a mixture of 20 ml. of 10 per cent. (by vol.) sulphuric acid saturated with potassium bromide, and 15 ml. of *n*-heptane. This

mixture is at once titrated with 0.5 *N* potassium bromide-bromate solution, until a faint yellow colour is obtained, and 1 ml. of the bromide-bromate solution in excess is then added. The solution is shaken for 2 minutes, and the iodine liberated on addition of 5 ml. of a saturated solution of potassium iodide is titrated with 0.1 *N* sodium thiosulphate solution, with starch as indicator. Then the bromine number = $(0.0799 \times \text{normality of } KBrO_3 \times \text{net vol. of } KBrO_3 \times 100)/\text{wt. of sample}$. This modified procedure eliminates the preliminary trial titration and the necessity for cooling the sample prior to titration found necessary by other workers (*loc. cit.*), thereby halving the time required for control tests. It is applicable to straight-chain olefines, diisobutylene and other branched-chain olefines, of low mol. wt., and is unaffected by the presence of pure aromatic hydrocarbons, although erratic results are obtained with highly-branched polymers and in presence of certain sulphur compounds; thus low concentrations of isoamyl mercaptan and higher concentrations of benzyl mercaptan, amyl mercaptan and isoamyl disulphide lower the bromine number of diisobutylene. The effects of these sulphur compounds can, however, be overcome by the use as a catalyst of 1 ml. of a 10 per cent. solution in water of mercuric chloride, zinc sulphate or uranium acetate, or of 0.1 g. of solid silver bromide. Data tabulated for a number of pure unsaturated aliphatic hydrocarbons, synthetic blends made from naphtha (A.S.T.M.), *n*-heptane, diisobutylene and heptene-1, and also for a number of saturated compounds, show satisfactory agreement with the theoretical values. (*Abstractor's Note*.—The solubility of mercuric chloride in cold water is less than 10 per cent.) J. G.

Isolation of Arachidonic Acid. G. Y. Shinowara and J. B. Brown. (*J. Biol. Chem.*, 1940, 134, 331-340.)—The phosphatides of the lipids of beef suprarenals were converted into the corresponding methyl esters by direct alcoholysis. The esters were dissolved in acetone, and the solution was cooled to -20° C., causing the saturated esters to crystallise out. These were filtered off, and the filtrate was cooled to -70° C.; the resulting crystals, containing most of the methyl oleate, were filtered off, and the filtrate, containing esters that were 50 to 60 per cent. methyl arachidonate was concentrated and cooled again to -70° C. The final filtrate contained 70 to 75 per cent. methyl arachidonate. By fractional distillation, the purity was raised to 90 to 95 per cent. An alternative method of purification was also used in which the impure material was brominated in ethereal solution at 0° C. and the resulting octabromides, m.p. 228.5 to 229.5°, were debrominated with zinc and boiling methyl alcohol. The two methods gave essentially the same substance, except that the second method yielded purer material with an appreciably lower polybromide value than the first method. The following con-

stants were found for methyl arachidonate prepared by the crystallisation-distillation and bromination-debromination methods respectively: n_D , 1.4800–1.4845, 1.4823; iodine value, 289.3–306.2, 312.8–319.0; polybromide value, 87.0–97.7, 86.3–86.6; thiocyanogen value, 159.0, 161.1. Arachidonic acid is known to be a straight chain eicosatetraenoic acid, $C_{20}H_{32}O_4$; the straight chain structure was confirmed by analytical data on its hydrogenation product. Ozonolysis experiments, designed to determine the position of the double bonds, indicated that two of these are probably in the 6 and 18 positions; the evidence is against the presence of double bonds in the 9 and 12 positions as in linolic and linolenic acid. There is some evidence from the diene number that the methyl arachidonate prepared by the debromination method contains 5 per cent. of conjugated double bonds, supporting the belief that this ester is a mixture of at least two compounds, one of which contains a conjugated double bond system. F. A. R.

Analyses of Certain South African Woods, with Special Reference to their use as Producer-Gas Generator Fuels. P. E. Hall. (*J. Chem. Met. Mining Soc. S. Africa*, 1940, 40, 350–352.)—The woods tested were (1) *Eucalyptus cladocalyx*, *E. saligna* and *E. paniculata*; (2) *Acacia karroo*, *A. saligna*, *A. cycloptis* and *A. mollissima*; (3) *Pinus insignis*, *P. pinaster*, *P. patula*, *P. longifolia* and *P. caribaea*. The most important properties in the evaluation of woods for producer-gas operation were found to be the following:—*Sp.gr.*—Segments cut from "rounds" of wood, 0.25 inch thick, were weighed in air before and after waxing, and then in water, the sp.gr. being calculated on the assumption that the volumes of the waxed and unwaxed woods were equal. The values for the above 3 groups of woods were 0.57 to 1.03, 0.68 to 0.86 and 0.38 to 0.65, respectively. A high sp.gr. is an advantage, because a vehicle must provide carrying capacity for its own fuel. *Ash content.*—Whilst none of the woods had a value exceeding 1 per cent., the light powdery nature of the ash is such that it will contaminate the gas made and so necessitate a relatively large area of filtration, a matter of importance with down-draught producers. *Charcoal yield.*—Values obtained (Gray-King low temperature assay) were 20.7 to 25.8, 20.9 to 25.3, and 22.6 to 28.7 per cent., respectively. Since most of the fuel gas is derived from the charcoal, a high value is an advantage. *Tar yields* (Gray-King) were 10.6 to 28.0, 13.5 to 18.7, and 15.9 to 19.8 per cent., respectively. Unless the producer is of the down-draught type (in which the tar is cracked in the fuel-bed) a high tar-content is a disadvantage, since it tends to gum up the fuel gas-line and to interfere with the filter, so that frequent stoppages for cleaning are required. The calorific values were 7,410 to 7,680, 7,360 to 7,740 and 7,530 to 8,470 B.Th.U. per lb., respectively. On the whole, there are no marked differences in suitability between the woods, but the above considera-

tions suggest that the *Eucalyptus* group should be satisfactory fuels by reason of their high sp.gr. and charcoal yield, low ash content, and reasonable tar-yield; *E. saligna* has a rather low sp.gr. and *E. paniculata* a high tar-yield and a low charcoal yield. Woods of the *Acacia* group should prove fairly satisfactory, since the charcoal, tar-yield and sp.gr. are themselves fairly satisfactory. The low sp.gr. of the *Pinus* woods is a disadvantage (especially with portable generators); this is offset to some extent by their high charcoal yields as compared with the other woods. J. G.

Species Identification of Wood and Wood Fibres. (*Tech. Assoc. Pulp and Paper Ind. Amer., Standards*, Suggested Method, T.8 sm-37, Dec. 1937, pp. 1–12.)—Paper samples are torn up, and a small piece is boiled with 1 per cent. sodium hydroxide solution for a few minutes to remove the sizing. The alkali is washed out, the residue is shaken well with water, a drop of the resulting suspension is placed on a microscope slide, and a cover-slip is applied. The fibres may be stained with a 1 per cent. solution of methylene blue, a drop of which is placed by the side of the cover-slip and drawn under it by means of absorbent paper. After 3 minutes the excess of dye is removed by irrigation with water in the same way. Wood samples are broken into pieces about 2 cm. long and 3 mm. wide, which are boiled well in water until all air is expelled. The water is then replaced by conc. nitric acid, to which is added powdered potassium chlorate, and boiling is resumed until bubbles are evolved. When the fragments are bleached the chips are washed and disintegrated into the fibrous state by vigorous shaking. Reference standards should be prepared in this way from authentic specimens of wood. An illustrated list of the diagnostic features of 30 American pulpwoods is provided, and this, together with 3 keys, are used to identify the wood (*cf. Maby, ANALYST*, 1932, 57, 2). Diagnostic features not referred to by Maby include the following:—Tyloses, which are white, colourless or sometimes dark-coloured cell-like intrusions, which partly or completely fill the vessels of many hardwoods. They usually have thin walls, which are rarely pitted, although thick profusely-pitted walls sometimes occur. Ray-flecks are the characteristic broad faces of the wood ray as seen on a radial section; they may not, however, always be conspicuous, by reason of their lack of height, colour contrast or lustre. Two types of ray cells are distinguished, *viz.* those with bordered pits (ray tracheid cells) and those with none (ray parenchyma cells); unlike the tracheids, which are dead cells, the latter remain alive so long as they are part of the sapwood. The ray cells are to a great extent removed from chemical pulps by screening, but they are useful for the identification of mechanical pulps. The spiral thickening of the cell wall also is characteristic of certain woods, and it may occur at the tips of the vessel segments, where vessels come into

contact, or throughout each vessel segment. The following principal diagnostic features are described for each tree listed in the key, and serve for its identification:—*Gross features*.—Colour, hardness, texture and density of sapwood and heartwood; odour; width and uniformity of distribution of rings; nature of transition from spring-wood to summer-wood; texture and size of rays; number, appearance and size of longitudinal and transverse resin canals and ray flecks. *Minute anatomy*.—Number and average diameter of tracheids; position, number, grouping and nature of bordered pits; appearance of ray tracheids, and nature of their walls; presence of thin-walled epithelial cells associated with resin canals; dimensions of longitudinal and transverse canals; and degree of homogeneity of rays. J. G.

Inorganic

Colorimetric Determination of Copper with Triethanolamine. J. H. Yoe and C. J. Barton. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 456–459.)—The colorimetric determination of copper was made spectrophotometrically. With triethanolamine, the sensitiveness is substantially the same as with ammonia, though slightly greater at low copper concentrations. Triethanolamine has no odour and is not subject to losses by volatilisation. Ammoniacal copper solutions obey Beer's law, giving a linear relation between concentration and transmission at $625m\mu$ over the range 0 to 1000 p.p.m. of copper in 0.3M and in 2.5M ammonia; solutions in triethanolamine do not entirely conform to Beer's law at various concentrations from 1 to 5 per cent., as the curves show a slightly greater slope with low copper concentrations (up to about 60 p.p.m.). A series of curves are given showing the results obtained with the two reagents. W. R. S.

Colorimetric Determination of Free Chlorine in Effluents. F. J. Hallinan. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 452–453.)—The method, which is applicable to coloured effluents, is based upon the formation of an insoluble adsorption compound of α -naphthoflavone and iodine which is filtered off and subsequently dissolved in alcoholic potassium iodide solution. The effluent (50 ml.) is treated with 1 g. of solid potassium iodide and a buffer (1 g. of potassium sodium tartrate for pH 6.7, ammonium acetate for pH 8.9, sodium diethylbarbiturate for pH 10.0), which are dissolved by stirring, and after 2 minutes with 0.5 ml. of 0.2 per cent. α -naphthoflavone solution in 95 per cent. alcohol. After 3 minutes' standing the liquid is vigorously stirred to promote flocculation and filtered by suction through hard paper in a Gooch crucible. The clear filtrate is tested for iodine with more of the reagent, and if a positive reaction is obtained a fresh test must be carried out on a smaller volume of effluent. The precipitate is washed with chlorine-free water and extracted with about 5 ml. of a

0.1 per cent. alcoholic solution of potassium iodide, and the yellow filtrate is matched against permanent colour standards corresponding with known concentrations of chlorine. The method is very delicate, 0.05 p.p.m. of chlorine giving a perceptible yellow tinge in the small volume of alcoholic filtrate. W. R. S.

Colorimetric Determination of Iron with Salicylaldoxime. D. E. Howe and M. G. Mellon. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 448–450.)—A weight of sample containing 0.01 to 1 mg. of iron is dissolved in 10 ml. of nitric acid (1 : 3), and the red fumes are boiled off. After cooling, 10 ml. of 3 per cent. hydrogen peroxide, a slight excess of ammonia, and dilute hydrochloric acid to faint acid reaction (litmus), are added. Any insoluble matter is filtered off and well washed. The filtrate is treated with 10 ml. of 0.1 per cent. salicylaldoxime solution and, after agitation, with 1 g. of ammonium acetate. The solution is diluted to 100 ml., and an aliquot part is transferred to a Nessler tube and compared with a scale of standards prepared in the same manner less than 24 hours before use. A Duboscq colorimeter may also be used, the solutions obeying Beer's law. The reagent is prepared from a weighed amount of salicylaldoxime dissolved in 5 ml. of alcohol and diluted with the required volume of water. The interfering ions are copper, nickel, cobalt, molybdenum, uranium, chromium, phosphate, fluoride, and borate. They should be absent or removed from the solution. W. R. S.

Phosphate Separation in Qualitative Analysis. J. N. Friend, R. H. Vallance and H. J. G. Challis. (*Nature*, 1940, 146, 63.)—Since zirconium phosphate is insoluble in acid solution, a soluble zirconium salt can be used in place of ferric chloride for the removal of phosphates after the separation of the Group II metals as sulphides in qualitative analysis. Hydrogen sulphide is removed from the Group II filtrate by boiling, in the usual way, and a few drops of nitric acid are added, followed by an acid solution of zirconium nitrate, a drop at a time, until precipitation is complete. The solution is warmed, and after separation of the precipitated zirconium phosphate the filtrate is tested with a drop of reagent to ensure that no phosphate remains. Ammonium chloride and ammonium hydroxide are then added, and the solution is boiled well (important) and filtered. The filtrate may contain the members of Groups IV, V and VI, and is dealt with in the usual way, whilst the precipitate contains any iron, chromium or aluminium and the excess of zirconium salt. Aluminium is separated by extraction with boiling sodium hydroxide solution, the extract is neutralised with acid, and the aluminium is precipitated with ammonia. The residue (which is brown if iron is present) is oxidised by addition of water and sodium peroxide, when any chromium passes into solution as chromate. A solution of any insoluble residue in acid is tested for the presence of iron, and a confirmatory

test for chromates should be applied to the filtrate because zirconium salts sometimes contain titanium, and this will have formed the coloured pertitanate, which will mask or simulate the colour of the chromate. The advantage of the method is that it is more straightforward than that usually adopted, whilst the disadvantage is the cost of pure zirconium nitrate. Zirconia, however, is relatively cheap, and the presence of hafnium in it is immaterial as it behaves like zirconium under the above conditions. The reagent is best prepared by boiling a solution of the nitrate with an excess of sodium hydroxide (to extract any aluminium), washing the precipitate repeatedly by decantation, and allowing it to settle overnight; it is then suspended in water and dissolved by the addition of half the volume of warm 50 per cent. nitric acid.

J. G.

Determination of Sulphides in Depilatories. E. M. Hoshall. (*J. Assoc. Off. Agr. Chem.*, 1940, 23, 437-444.)—Liquid depilatories contain from 2 to 10 per cent. of alkali sulphides calculated as the anhydrous salts. Powder depilatories contain 20 to 30 per cent. of barium or strontium sulphides, and mixtures of zinc sulphide and lime have also been used. Paste preparations form the largest class, and contain 5 to 20 per cent. of barium or strontium sulphide and occasionally calcium hydrosulphide and saccharosulphide. Methods for the determination of the sulphide-content of these preparations were examined, and many were found too tedious and cumbersome for routine purposes, but the following modification of Mohr's method gave satisfactory results:—An amount of depilatory equivalent to less than 0.12 g. of hydrogen sulphide is added to 50 ml. of 0.1N arsenious oxide solution, and the mixture is acidified with 20 ml. of dilute sulphuric acid (1 + 1) and shaken vigorously in a closed flask. If the depilatory contains much carbonate the flask is closed by means of a stopper carrying a separating funnel with a stem reaching nearly to the surface of the liquid, and the acid, mixed with 10 ml. of the arsenious oxide solution, is introduced gradually from the separating funnel. Traces of hydrogen sulphide and the liberated carbon dioxide bubble up through the acid, and the carbon dioxide can be removed by gentle shaking. When the reaction has stopped the contents of the funnel are rinsed into the flask. When cold the mixture is diluted to 250 ml. and filtered, the first 20 ml. of the filtrate being rejected. A portion (100 ml.) of the filtrate is treated with starch solution, sufficient 0.1N iodine solution to produce a blue colour, and 1 to 2 g. of sodium bicarbonate in excess of the amount required for neutralisation and titrated with 0.1N iodine solution. One ml. of 0.1N arsenious oxide solution is equivalent to 0.002556 g. of hydrogen sulphide. The method was found to be sufficiently accurate for routine purposes, and no substances interfering with the reaction were encountered. Discrepancies in the

results, when they occurred, were found to be due to instability of the sulphides (particularly sodium sulphide) in presence of other constituents of the depilatory. With preparations that carbonise or dissolve only with difficulty, 15 ml. of hydrochloric acid may be used instead of dilute sulphuric acid.

A. O. J.

Microchemical

Permanganate Acid Ashing Micro-method for Iodine Determinations. I. Values in Blood of Normal Subjects. D. S. Riggs and E. B. Man. (*J. Biol. Chem.*, 1940, 134, 193-211.)—The various methods hitherto used for the estimation of iodine in blood are subject to certain errors inherent in each method, and an entirely new method of oxidising the organic matter has been introduced, and is claimed to avoid these errors. Ten ml. of the oxalated blood are oxidised with potassium permanganate and dilute sulphuric acid, ceric sulphate and copper being used as catalysts. The iodine is removed by distillation with oxalic acid, and the iodine in the distillate is oxidised to iodate by potassium permanganate and then determined by titration with sodium thiosulphate. **Apparatus.**—The distillation apparatus consists of (1) a 900-ml. Kjeldahl flask fitted with a ground-glass joint; (2) a distilling arm, made of wide glass tubing bent twice at right-angles, with a ground-glass joint at each end, one of which fits the Kjeldahl flask. Sealed through one bend of the distilling-arm is a dropping funnel, the stem of which is made of capillary tubing; the capillary extends to within 8 mm. of the bottom of the flask; (3) a spiral condenser, 17 cm. long, with a ground-glass joint into which the other end of the distilling-arm fits; (4) a 250-ml. wide-mouthed Erlenmeyer flask with a short horizontal side-arm sealed on at the base; this tube has a capacity of 3 ml. and is calibrated at 2 ml.; (5) a thermometer (120 to 200° C.) attached to the capillary tube of the distilling arm by strong rubber bands. **Digestion.**—Into the digestion flask are introduced 14 g. of potassium permanganate (recrystallised from water), a small piece of copper, 10 mg. of ceric sulphate (washed twice with 5 times its weight of boiling 95 per cent. alcohol) and 15 to 25 ml. of redistilled water. While the mixture is shaken, 10 ml. of oxalated blood are added, and then 10 ml. of 18N sulphuric acid, and the flask is again shaken. A violent reaction takes place, and after about 5 minutes, when it has subsided, a further 200 ml. of 18N sulphuric acid are added. The flask is heated cautiously over a wire gauze until the contents are boiling steadily, digestion being continued until a temperature of 195° C. is reached; the solution is then allowed to cool to 100° C., 150 ml. of 1 per cent. potassium permanganate solution are added, the flask is heated to 145° C. for about 15 minutes and again allowed to cool to 100° C., and the thermometer is washed with 25 ml. of water and removed. **Distillation.**—The digestion

flask is connected to the distillation apparatus, and the receiving flask, containing 1 ml. of *M* potassium carbonate solution and 1 ml. of 0.1 *M* sodium bisulphite solution, is tilted so that the tip of the condenser dips below the surface of the liquid. The digestion flask is heated to 138 to 140° C. and oxalic acid solution (saturated at 30° C.) is added slowly through the dropping-funnel. Two or 3 ml. are added beyond the point at which the solution is decolorised, followed by water so long as the distillation is continued. About 170 ml. of distillate are collected in 30 minutes. The distillate in the receiving flask is evaporated cautiously on an electric hot-plate to a volume of 6 or 7 ml. **Oxidation.**—The flask is placed in a water-bath maintained at 70 to 80° C., and 8 drops of 0.2 *M* potassium permanganate solution are added. If these are decolorised, more permanganate is added until a purple colour persists. After 4 minutes 10 drops of 8 *N* sulphuric acid are added, and after a further 4 minutes the flask is removed from the water-bath and 0.75 *N* sodium nitrite solution is added dropwise, with constant shaking, until the solution is quite clear. One drop of nitrite solution is added in excess, followed immediately by 2 drops of 5 *M* urea solution. The flask is replaced in the water-bath until the volume is about 2 ml. (measured with the aid of the side arm). **Titration.**—The flask is cooled in ice-water and a drop of 1 per cent. stabilised starch solution and then 0.06 ml. of freshly prepared 0.2 per cent. potassium iodide solution are added. The solution is titrated with 0.001 *N* sodium thiosulphate solution delivered from a micro-burette, the tip of which dips just under the surface of the liquid being titrated. **Blank.**—A blank is prepared by the method used in the estimation itself, with the following modifications: 15.5 g. of permanganate are used for the oxidation, anticoagulant equal to the amount present in the blood is introduced into the digestion flask, 85 ml. of water are used to wash the thermometer, and, before oxidation of the 7 ml. of distillate with permanganate, a small, accurately measured quantity (0.5 ml. of 0.00005 *N*) of biiodate solution is added. A control titration is carried out on the same amount of iodate added to a mixture of 1 ml. of 0.1 *M* sodium bisulphite solution and 1 ml. of *M* potassium carbonate solution and carried through the final oxidation and titration processes. The value of the blank is the difference between the titres of the control iodate samples and of the distillate to which iodate was added. **Results.**—In 59 experiments inorganic and organic iodine was recovered, with an average error of 6.5 per cent.; the average recovery for all experiments was 95 per cent. F. A. R.

Micro-determination of Succinic Acid. G. J. Goepfert. (*Biochem. J.*, 1940, **34**, 1012-1014.)—The following method for the estimation of minute amounts of succinic acid in nutrient media containing nitrate, sulphate and phosphate ions was devised. The solution, containing volatile acids and 2 ml. of 0.1 *N*

succinic acid solution, was acidified with conc. sulphuric acid until acid to Congo red, heated on the water-bath, and treated with 0.1 *N* potassium permanganate solution until a brown precipitate was formed. This was dissolved by addition of sodium sulphite, and the solution was evaporated to dryness. The residue was dissolved in 15 ml. of chloride-free water, 2 ml. of conc. sulphuric acid were added, and the solution was saturated with potassium sulphate and extracted with ether for 3 or 4 hours in a continuous extractor. To the extract were added 5 ml. of chloride-free water, and the ether was removed by distillation. The aqueous residue was boiled over a flame for 30 seconds and cooled under the tap, and one drop of *m*-nitrophenol indicator (0.3 per cent. in water) was added, followed by 0.05 *N* sodium hydroxide solution until one drop caused the solution to turn yellow. It was decolorised with one drop of 0.1 *N* nitric acid, and a measured excess of 0.02 *M* silver nitrate solution was added immediately. The solution was made neutral by adding one drop of 0.05 *N* ammonia and allowed to stand in the dark for 2 hours. The precipitate was collected on a Gooch or sintered-glass crucible and washed successively with 3-, 3- and 2-ml. portions of 1 per cent. ammonium nitrate solution. The filtrate was treated with 2 drops of dichloro-fluorescein indicator (0.1 per cent. in 70 per cent. ethyl alcohol) and about 8 drops of 1 per cent. soluble starch solution (chloride-free). The excess silver was titrated with 0.02 *M* potassium bromide solution until the pink colour disappeared. (One ml. of 0.02 *M* silver nitrate solution = 1.18 mg. of succinic acid.) In 25 samples the recovery was 98 ± 2 per cent. F. A. R.

Micro-estimation of Uronic Acids. E. M. Kapp. (*J. Biol. Chem.*, 1940, **134**, 143-150.)—When glucuronic acid is boiled in hydrochloric acid in presence of naphthoresorcinol, a purple ether-soluble pigment is formed (Tollens' reaction). The usual conditions employed for this reaction are not optimal, however, and an improved method, suitable for quantitative purposes, has therefore been devised: A sample containing 10 to 40 γ of free glucuronic acid (or 7 to 30 γ of galacturonic acid) in 3.5 ml. of water is treated in a 10- or 15-ml. centrifuge tube with exactly 0.1 ml. of a 1 per cent. solution of naphthoresorcinol in 95 per cent. ethyl alcohol and then with 1.5 ml. of conc. hydrochloric acid. After mixing, the tube is immersed in a boiling-water bath for 4.5 hours with occasional shaking (2 hours is sufficient for galacturonic acid), cooled and centrifuged. Most of the supernatant liquid is withdrawn by means of a pipette and 10 volumes of water are added to the remainder. The resulting aqueous suspension is extracted several times with 1-ml. portions of ether, and the combined extracts, measuring not more than 6 ml., are dried with sodium sulphate and filtered into a 10-ml. flask. The solution is made up to the mark, and the colour is measured as soon as possible in a step-photometer with filter S 57 and

20 mm. cells, against an ether blank. The colour produced by the reagents alone is measured, and the value so found is subtracted from the value given by the sample. The amount of glucuronic or galacturonic acid is calculated from a standard curve obtained with pure specimens, or from the two equations: Mg. of glucuronic acid = $(D - 0.08)/18.7$, and Mg. of galacturonic acid = $(D - 0.08)/24.5$, where D is the observed photometric density.

The two substances are readily distinguished from one another by the rate of colour development, the maxima for galacturonic and glucuronic acid being reached in 2 and 4.5 hours respectively. A number of substances, including pentoses, interfere with the colour, and must be removed before carrying out the reaction. The agreement between the amount of glucuronic acid added to previously analysed specimens of urine and the amount found to be present was good, the largest discrepancy being 3 γ in 39 γ . F. A. R.

Physical Methods, Apparatus etc.

Estimation of *o*-Nitrophenol in *p*-Nitrophenol, and *o*-Aminophenol in *p*-Aminophenol by Fluorescence Analysis. W. Seaman, A. R. Norton and O. E. Sundberg. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 403-405.)—The reaction depends on the fact that *o*-aminophenol (e.g. produced by reduction of *o*-nitrophenol) reacts with carboxylic acids or their anhydrides to form benzoxazole and associated fluorescent by-products, whereas *m*- and *p*-aminophenols do not. A quantity of sample equivalent to 1.3 g. of nitrophenol is boiled under a reflux condenser with 125 ml. of water, 30 ml. of conc. hydrochloric acid and 5 g. of zinc dust for 15 minutes. The mixture is cooled and filtered, the residue is washed with three 10-ml. portions of water, and the total filtrate is treated with 15 ml. of ammonia, followed by sufficient 2 per cent. ammonia to bring the pH to about 5.1 (i.e. alkaline to Congo red paper and faintly alkaline to red litmus paper); this ensures that no zinc hydroxide is precipitated. Most, or all, of the *o*-aminophenol but little of the *p*-aminophenol may then be removed from the cold solution by extraction with three 25-ml. portions of ether; these are united, washed with two 5-ml. portions of water, and evaporated rapidly just to dryness, the formation of an adherent skin being avoided so far as possible. The residue is mixed with 5 g. of sublimed benzoic acid without delay (to avoid oxidation), and the mixture is heated in an oil-bath at 155° to 160° C. for 15 minutes after the contents have melted; carbonisation should be avoided. The cold residue is rubbed alternately in ammonia and in water until no more will dissolve, and the resulting blue solution is extracted with two 25-ml. portions of benzene, which are then mixed and extracted, first with 10-ml. portions of 1 per cent. sodium hydroxide solution until

no more blue colour is removed, and then with water until the washings are neutral to litmus. The fluorescence of the benzene solution is matched in a test-tube (150 \times 25 mm.) against a series of standards prepared from known mixtures of pure *p*-nitrophenol with 0 to 0.5 per cent. of *o*-nitrophenol. A fairly good comparison may be made with the aid of an ordinary desk-lamp, if the eyes are shielded against direct light and the tubes are placed vertically on a dark background and illuminated from the side and viewed from above. A more sensitive comparison may be made in filtered ultra-violet light in a dark room, the tubes being held in a rack at arm's length, slightly above eye-level over the lamp. The minimum detectable quantity of *o*-nitrophenol in *p*-nitrophenol is 0.05 per cent., and although no mixtures containing *m*-nitrophenol were used, it was shown that *m*-aminophenol gives no fluorescence. Any red colour obtained with commercial samples of *p*-nitrophenol is eliminated in this way, although the effect of the red colour obtained when more than 1.3 g. of sample is used, or when the method is used to estimate commercial *o*-aminophenol (the reduction process being omitted) cannot then be avoided. The following modified technique should then be used. A solution of 1 g. of sample in a mixture of 100 ml. of water and 3 ml. of conc. hydrochloric acid is filtered, and the above procedure followed from the stage of the adjustment of the pH value. The final benzene extract is shaken first with 50 per cent. ammonia (twice), and then with 10-ml. portions of 33 per cent. hydrochloric acid until no further colour is removed. If the solution is then filtered through a dry paper, it may be matched in the usual way without any appreciable loss of fluorescence; if desired, the standards may be treated similarly. A solution in benzene of phenyl benzoxazole prepared by the reaction of *o*-aminophenol with benzoyl chloride had a red-brown fluorescence, but successive recrystallisations from acetone and a petroleum hexane cut converted this into a green fluorescence, which ultimately disappeared once the correct m.p. had been attained and had not reappeared after 16 months. It is concluded that the fluorescence matched is due to a by-product of the benzoxazole condensation. J. G.

Quantitative Analysis of Plant Tissues for Lithium by the Ramage Flame Spectrographic Method. N. L. Kent. (*J. Soc. Chem. Ind.*, 1940, 59, 148-153r.)—The weighed sample is dried at 100° C., and ashed, and about 20 mg. of the ash are weighed into a small specimen tube and dissolved in a volume of dilute nitric acid (1:10) such that 0.1 ml. of solution contains exactly 0.25, 0.5, 1 or 2 mg. of ash, according to the amount of lithium believed to be present. Then 0.1-ml. portions of the solution are dropped on to quarter-circles of filter paper (No. 60, 9 cm.), which are dried on a glass plate over a 60-watt lamp and rolled into tight spirals about 1.75 in. long. The spiral is then "fed" horizontally into the

hottest part of the flame of a (Hilger) Ramage oxy-coal gas burner (*cf.* Steward and Harrison, *Ann. Bot.*, 1939, NS3, 427) at the rate of about 2 in. per 10 seconds, and the resulting spectrum is photographed by means of a Hilger Medium Quartz Spectrograph, E3, panchromatic plates (Ilford process, 10×4 in.) being used, since the most sensitive lithium lines are in the red at $670.8 m\mu$. A novel feature of the present apparatus is the inexpensive feeder. This consists of a horizontal piston, into which the paper "spiral" can be fixed in such a way that when a large hand flywheel is turned the piston moves forward, thereby enabling the tip of the spiral to be advanced rapidly through the relatively cool outer cone of flame, until it begins to burn brightly at the edge of the inner cone formed by the junction of the oxygen and coal gas. Manipulation of the flywheel enables the spiral to be maintained in this position, and a hinge serves to swing the piston in and out of position for loading and removal of the ash; 50 exposures can be made in 2 hours, 25 spectrograms being recorded at each end of the plate, which is developed in metol-quinol at 20°C . for 3.5 minutes and fixed in acid "hypo" for 15 minutes. The standard spectrograms used for comparison purposes are prepared in a similar way from solutions of lithium sulphate equivalent to 1 mg. to $0.05 \mu\text{g}$. of lithium per 0.1 ml., the maximum sensitiveness of the method being obtained with quantities between 0.4 and $4.0 \mu\text{g}$. It is therefore sometimes necessary to make a preliminary trial analysis in order to ensure that this condition will be satisfied in the final analysis. The maximum probable error, as estimated from the errors of the individual operations, is 9 per cent. The objects of the analyses were to ascertain how much lithium is taken up by plants from normal soils and from soils treated with lithium chloride or nitrate, and any resulting differences in the distribution, concentration and uptake of the lithium in the plant. With each species of plant examined (*viz.* healthy wheat and celery plants, tomato crown galls dissected from the stems of plants that had been inoculated with *B. tumefaciens* and treated with various amounts of lithium nitrate, and the dead leaves which fell from these plants), there was a high degree of correlation between the concentrations of lithium in the soil and in the plant. The greatest accumulations in wheat were found in the oldest leaves, and in celery in the margin of the largest leaves. The tolerance for lithium was in the decreasing order, wheat, celery and tomato. Plants appear to have little power to prevent the absorption of lithium, and it is suggested that the accumulation of lithium in the leaves depends more on their growth-rates than on transpiration; lithium performs no useful function, and merely accelerates leaf-fall. The following are the lithium-contents (in mg. per 100 g. of fresh material) obtained for plants grown on untreated soil:—Wheat leaves, 2.81, roots, 1.73; celery leaves, 1.5, petioles, 0.09, roots, 0.28; tomato petioles, 0.26. The corresponding figures for the

lithiated plants having the maximum individual lithium-contents were, 33.8, 36.5, 29.8, 1.66, 4.08, and 1.49, respectively. J. G.

Standard Method for Testing the Fastness to Light of Coloured Textile Materials. L. A. Lantz. (*J. Tex. Inst.*, 1940, 31, S13-14.)—The results obtained by the method proposed in the Report of the Society of Dyers and Colourists (*ANALYST*, 1934, 59, 783) were compared with those obtained by the methods suggested by other organisations, with the object of agreeing on a common range of standards which might be adopted internationally. An improved set of Tentative Standards dyed on wool* are, therefore, now submitted for publication in accordance with the agreements made by the Textile Institute with the Society and with the British Standards Institution; these are:—(1) 0.8 per cent. Brilliant Wool Blue FFR extra (I.G.); (2) 1.0 per cent. Brilliant Wool Blue FFB extra (I.G.); (3) 1.2 per cent. Brilliant Indocyanine 6B (I.G.); (4) 1.1 per cent. Polar Blue G conc. (Gy.); (5) 0.8 per cent. Solway Blue RS (I.C.I.); (6) 2.5 per cent. Alizarine Light Blue 4GL (S.); (7) 2.5 per cent. Soledon Blue 4BC pdr. (I.C.I.); (8) 3.0 per cent. Indigosol Blue AGG (I.G.). The parallel red series formerly suggested has been abandoned as superfluous. Appropriate adjacent members of this series are exposed together with the material to be tested until the latter shows definite signs of fading; its fastness number is then that of the Standard which has faded to the same extent. The standards are little affected by variations in atmospheric humidity and temperature, and they allow reliable figures to be obtained under all conditions of daylight and in different climates. If an artificial source of light is used it should approximate to daylight as closely as possible; when it is an enclosed carbon arc, provision should be made for the control of the humidity and temperature of the atmosphere surrounding the samples, which should be disposed in a circle around the arc on a rotating device, so that the distribution of the light falling on them is equalised. When fastness measurements are based on times of exposure to a constant source of light (as distinct from comparisons with a series of standards) the suggested Standards may be used to control the exposures and to calibrate the lamp used. J. G.

Microscopical Investigation of True Woollen Cloths. Chemical and Microscopical Determination of Additions of Casein Wool. J. Straub and G. J. Van Amerongen. (*Chem. Weekblad*, 1940, 37, 236-238.)—For the determination of the proportions of damaged wool in cloth used for military uniforms, a thread from the cloth is cut into a piece 0.5 to 1 mm. long and mounted in a drop of 50 per cent. glycerin or a mixture of glycerin and gelatin on a microscope slide

* Obtainable from the Society of Dyers and Colourists, in sets, $4\frac{1}{2} \times 3\frac{1}{2}$; price, 5s. per set.

and covered with a cover-glass on which is etched a scale which enables the widths of the fibres to be estimated. A table shows the relationship between the thicknesses and the colours of undamaged and damaged wool as seen under the microscope. Most of the fibres of 0 to 15μ in thickness were white; those of 15 to 30μ were white and either blue-green or yellow-green, respectively; those over 30μ were black and blue-green or yellow-green, respectively. The number of fibres falling in these three thickness groups were, respectively, 7, 28 and 0 for the undamaged wool and 5, 25 and 7 for the damaged wool. Estimations of the proportions of undamaged fibres based on the frequency of distribution of the thicknesses and colours observed gave closely concordant results. The magnifications used for these counts were $60\times$, but the details of the differences between damaged and undamaged wool were shown better at magnifications of $130\times$. Casein wool is recognisable by its comparatively thin uniform fibres, with their narrow central canal, and the absence of scalariform markings (magnification, $130\times$). Photomicrographs illustrate the differences between the three types of fibre. The average weights per cm. of length of true wool and casein wool from a sample of cloth were (after correction for the effects of mechanical treatment) 9.1 to 5.1y, respectively, and these figures were used to calculate the weights of each type of wool present in the cloth from the microscopical counts obtained with 3 threads

each from the warp and weft. In the chemical method described (*cf.* Herzog, *Technologie der Wolle*, 1938) 0.4 g. of the sample is dissolved in a 40-ml. porcelain crucible in 10 ml. of 20 per cent. hydrochloric acid, or in a mixture of sodium hydroxide solution and bromine water (*cf.* Trotman and Bell, *J. Soc. Chem. Ind.*, 1926, 45, 10; *ANALYST*, 1934, 59, 715). After 30 to 60 minutes 1 ml. of a solution containing 25 g. each of cupric nitrate and sodium chloride and 10 g. of sodium nitrate in 100 ml. of water (Benedict-Denis reagent) are added, and the mixture is evaporated to dryness on the water-bath. Evaporation is repeated after a further addition of 4 ml. of reagent, and the residue is ignited in a muffle at a glowing heat for 3 minutes with the crucible covered, and then for 10 minutes with it open. The sulphate-content of a solution of the residue in 10 ml. of 10 per cent. hydrochloric acid and 10 ml. of water is then determined by precipitation as barium sulphate, and the sulphur-content of the original sample may be calculated from this. The average sulphur-contents of the true wool and casein wool in the cloth under examination were found to be 2.81 and 0.43 per cent., respectively, and these data enabled the proportions of the two types of fibre in the cloth to be calculated from its sulphur-content. The ratios of casein wool to true wool in the cloth, as found by the chemical and microscopical methods, were 29 : 71 and 28 : 72 respectively, and as certified by the manufacturers 30 : 70. J. G.

Reviews

THORPE'S DICTIONARY OF APPLIED CHEMISTRY. Fourth Edition. By Sir J. F. THORPE, C.B.E., D.Sc., F.I.C., and M. A. WHITELEY, O.B.E., D.Sc., F.I.C. Vol. IV. Pp. 603. London: Longmans, Green & Co. Price 70s.

This volume was published on June 10th of the present year. On the same day, with tragic suddenness, Prof. Sir Jocelyn Thorpe peacefully passed away in his home in Sussex. Appreciations of Thorpe and tributes to his genius and geniality have appeared in many places; to these one must add that, since the mantle of Sir Edward Thorpe fell upon him, he has by this Dictionary made a memorial for himself which will cause his name to be remembered (with that of Sir Edward) with gratitude by the rising generation who could not know him so well as those who are older.

It may perhaps be impossible to make Thorpe's Dictionary quite the same without "Thorpe," but the framework exists and Dr. Whiteley has the co-operation of many well-known experts as contributors.

The present volume ranges from Digallic Acid to Feeding Stuffs; it differs a little from the preceding three in that it contains perhaps a smaller number of headings, but certainly a greater number of what may be called major articles. Even these are too numerous to mention in a brief review; some of them amount almost to small text-books on their subjects. For example, the articles on Explosives, including Gaseous Explosion and Explosives used in Coal Mines, occupy no less than 146 pages and vie with certain well-known textbooks in their comprehensiveness of survey in this field. Although it is probably true that no

new major explosives have been introduced lately, the many modifications—both of production and of product—are of great importance; these are here clearly but concisely described. There are, too, some major articles of particular interest to the laboratory worker, such as those on laboratory distillation, laboratory extraction apparatus and on disinfectants. The articles on azo dyestuffs and on dyeing (48 and 68 pp. respectively) are full of interest to the general chemist; including the analyst. The classification and exposition of the constitution of azo dyes in terms of function is interesting and, so far as the Dictionary is concerned, novel. This method conveys a picture of the structure of these interesting compounds more readily than conventional though fuller formulae. There is some overlapping between the articles on dyestuffs and dyeing, but this is perhaps inevitable, and we think that out of a 68-page article on dyeing, one-quarter page is not a wholly adequate allowance for the complexities of modern fur dyeing.

As in previous volumes, there is a nice balance between matters industrial and matters theoretical, and it is very satisfactory that the authors maintain the principle of setting out clearly the scientific bases of the subjects treated as well as the practical industrial applications.

In perusing the list of contributors it is as pleasant to welcome new names as it is gratifying to see some old and much-honoured contributors, among whom may be mentioned Dr. Bernard Dyer who has revised and brought up-to-date his contribution on feeding stuffs. The general usefulness of the volume is such that it becomes indispensable to the consulting chemist, who naturally turns to it in all his problems. Even the expert in a particular field finds much in it that is helpful. We are sorry to note the increase in price, as the purchase of these volumes is already a quite considerable item of expenditure, but we suppose in these days it is unavoidable. These are volumes which people who use such books must acquire, more or less regardless of cost.

H. E. Cox

INORGANIC CHEMISTRY. FRITZ EPHRAIM. Third English Edition. By P. C. L. THORNE, M.A., M.Sc., Ph.D., F.I.C., and A. M. WARD, Ph.D., D.Sc., F.I.C. Pp. xii + 911. London: Gurney & Jackson. 1939. Price 28s.

An organic chemist, surveying the achievements of inorganic chemistry up to date in, say, the year 1895, might well ask (in fact often did ask) the question put to Ezekiel in the valley of dry bones—"Can these bones live?" The inorganic chemist of that day, less cautious than the prophet, was very apt to reply, "No they can't; you will have to take them as they are." The coming together of the bones, the clothing them with flesh and skin, in some instances even the breathing into them of the breath of life, has been very largely a miracle of this century. The process is very far from complete; in some regions the bones seem hardly to have stirred, but in a surprisingly large number they are giving every indication of being alive. It makes a fascinating story—this achievement—and nowhere has it been better presented than in the book under review; it is more, not less, fascinating by very reason of the fact that it is incomplete.

The distinctly unconventional method of approach of former editions has been preserved; in the words of the preface, "The materials of inorganic chemistry are dealt with collectively rather than individually." To describe this method adequately is impossible; perhaps the nearest approach one can make is that the basis of classification tends to be anionic rather than cationic as in ordinary textbooks. Covering, as it does, however, the whole wide sweep of a subject so liable to change, not only in detail but even in fairly basic conception, it is necessarily, to a large extent, a new book. The afore-mentioned method of approach is undeniably stimulating; it has many and obvious virtues, among them being the cross-sections it gives of information to be found in other textbooks; it has also drawbacks which, perhaps, are not quite so obvious. The book is such a mine of valuable information that one needs must want not only to read and study it, but also to

use it as a book of reference; here, however, one comes up against the unconventional arrangement. The unfamiliar setting has removed those landmarks which, though their use may be subconscious, serve as pointers in most textbooks, and one is left with a singularly lost feeling if what is wanted does not appear in the index. It is difficult to think of a remedy for this; the index contains some 4100 references, and to provide one that would meet all requirements would be a labour of Hercules.

It is a thankless task looking for flaws in such a really outstanding work, but there are a few statements scattered through its pages which cannot be allowed to go unchallenged. For instance, on p. 453, "Solutions of . . . ferric salts to which tartaric acid has been added, are not precipitated by . . . ammonium sulphide." This statement, if true, would make invalid one of the major separations of mineral analysis. Again (p. 752), "Secondary alkali monofluophosphates are neutral to phenolphthalein but acid to methyl orange," and (p. 844), "Tempered steel may contain *more than 90 per cent.* of this carbide" (Fe_3C).

These, and a few others of the same sort, must surely be proof-reading slips, although it is hard sometimes to see what can have been intended. As has been said, these lapses are very few, but their occurrence is most regrettable, inasmuch as they shake confidence in the many statements which cannot be so readily checked. Nevertheless, when one considers the immense labour that must have gone to the mere compiling of material for this wonderful book, criticisms such as these shrink into insignificance. The volume is well bound and well printed on good paper; misprints exist but are very few indeed; proof-reading slips are more numerous but still few. An unusual and attractive feature is a brief appendix giving aid to the younger chemist in looking up literature before embarking on a line of research. It is both impossible and unnecessary, however, to enumerate and comment on the various excellent sections that this work contains; the fundamental thing about it is its unity. Through it, more than through any book I know, one sees the picture, dim as yet and blurred, of that which, grasped by the alchemists by an act of faith but denied until recently by science for lack of sufficient evidence, is now emerging into the light of day and growing steadily clearer as the years go by—the unity underlying the bewildering phenomena of inorganic chemistry.

B. S. EVANS

PHYSICAL CONSTANTS OF HYDROCARBONS. Volume II. G. EGLOFF. Pp. 605.
New York: Reinhold Publishing Co.; London: Chapman & Hall. 1940.
Price 72s. net.

It will not be necessary to introduce Volume II of Dr. Egloff's well-known work to those who are familiar with Volume I. The second of this four-volume series is the result of a survey of data available on the physical constants of the non-aromatic cyclic hydrocarbons. The aliphatics were covered by the first volume.

For each hydrocarbon the carbon skeleton and the available data on melting and boiling points, specific gravity and refractive index are given. The text is conveniently arranged so that new experimental results can be added if desired. Every figure shown bears its own bibliographical reference. There is a short introduction in which the author discusses the structure of the alicyclic hydrocarbons and defines his nomenclature.

The cyclanes (cycloparaffins or naphthenes may be more familiar titles for some) have perhaps lagged behind other hydrocarbons in their application to chemical industry. Most people will agree with Egloff's opinion that a new chemical industry could well be developed based on cyclane chemistry. The almost unbelievable progress made recently in petroleum technology, of which no small amount is due to Egloff, allows hydrocarbons to be broken down and built up differently almost according to plan. These developments, coupled with the vast amount of cyclanes in nature (500,000,000 barrels from crude petroleum alone

during 1939 according to Egloff's estimate), must increase the importance of these hydrocarbons.

Although the volumes in this series find their most obvious application in oil research laboratories, workers in many other fields will find them invaluable—in fact, it is not difficult to predict that generations of chemists yet unborn will one day find themselves indebted to Egloff for the service he is now performing.

P. N. FULLER

THE MANUFACTURE OF COMPRESSED YEAST. By F. G. WALTER, A.A.C.I.
Pp. viii + 254, with 29 figures, including 3 plates. London: Chapman & Hall, Ltd. 1940. Price 15s. net.

This volume is intended to provide a comprehensive account of the manufacture of yeast on an industrial scale, from the underlying scientific principles involved to the examination and packing of the finished product. The contents include a chapter entitled "The Yeasts, etc.," in which descriptions are given of alcoholic fermentation, the common varieties of yeasts, moulds, bacteria, enzymes, various cereals with other sources of starch, and the carbohydrates. This is followed by sections devoted to the preparation of nutrient media, the isolation of pure yeast cultures and the production of seed yeast, whilst the four succeeding chapters describe in detail the manufacture of yeast on an industrial scale from cereals, molasses, glucose and spirit fermentations by the differential and continuous processes. A chapter on the drying of yeast and yeast nutrients in panary fermentations is followed by the final one providing details of the design, construction and operation of the yeast factory, laboratory equipment, the determination of pH values, and methods of analysis of the raw materials and the finished yeast.

It is evident that the author is far more familiar with operations of the yeast factory than with the more scientific part of the subject, for the few errors which occur are confined almost entirely to the first two chapters. Thus the yeast vacuole does not consist of glycogen, as stated on p. 1; cassava juice is not mainly hydrocyanic acid, nor are the granules of cassava starch far smaller than those of any other starch-producing plant (p. 29); the term "inversion" is not appropriate to the conversion of starch into dextrin, maltose, etc., in the mash tun (p. 65), and ammonia cannot be directly titrated with *N* sodium hydroxide solution even by using methyl orange indicator (p. 90). In addition, several typographical errors occur throughout the text, although these are not serious.

The larger portion of the book, dealing with the design, construction and operation of the yeast manufacturing plant is excellent and thoroughly comprehensive, its value being enhanced by the numerous tables and charts of operation given together with frequent references to the necessity for biological cleanliness throughout. In this section the only item that invites criticism is the undue number of buffer solutions, ranging from pH 2.0 to 5.4, for use with methyl orange as the sole indicator.

The binding, printing and illustrations are excellent, but the index, although fairly satisfactory, requires revision. Not only are some items in the text omitted, but numerous entries are included under their adjective, verb, and so forth instead of under their principal noun. This work contains much valuable information, and, with the elimination of the errors referred to above, would be an admirable textbook and reference manual for all interested in the industrial manufacture of yeast, which is being used to an ever-increasing extent in animal and human dietary and in therapeutic practice.

T. J. WARD

THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

THE following candidates have been elected members of the Society:

Cyril Dickinson, B.Sc. (Vict. and Leeds), F.I.C., Public Analyst and Bacteriologist.

Arthur Geoffrey Grimwade, M.A. (Cantab.), A.I.C., Chemist in Government Department.

Ernest Avery Roff, B.Sc. (Lond.), Chief Analyst and Research Chemist with Chemical Manufacturers.

Samuel John Rowland, B.Sc., Ph.D. (Reading), A.I.C., Research and Analytical Chemist at Dairying Research Institute.

Franklin Clermont Scott, B.Sc. (Lond.), A.I.C., Analytical Chemist with Chemical Manufacturers.

Herbert Lawton Wright, Chief Chemist at Beet Sugar Factory.

Death

WE regret to record the death, on August 26th, of
Albert John Murphy,
who had been a member of the Society for forty years.

The Spectrographic Analysis of Tobacco Ash

BY F. G. H. TATE, F.I.C., AND H. KENNETH WHALLEY, B.A., B.Sc., PH.D.

ALTHOUGH much work has been devoted to the constituents of tobacco such as sugars, proteins, water, nicotine and total ash,¹ and to the influence of soil conditions upon the proportions of these constituents,² the systematic inorganic analysis of tobacco ash appears to have received little attention. Further, the determination of the area or country of origin of tobacco by chemical or other means has presented a problem for which no satisfactory solution has yet been found.

In the discussion of a paper,³ and subsequently, one of us suggested that spectrographic methods of analysis might be of use in attempting the differentiation of sources of origin of tobacco. Following this suggestion, analyses have been made of over 300 samples of tobacco ash, involving more than 3000 quantitative

and partly quantitative determinations. Evidence has thereby been accumulated which not only supplies new data on tobacco constituents but also indicates the lines upon which discrimination between certain different types of tobacco can be made with a high probability of success. Preliminary work was carried out by arcing a 50/50 mixture of the ash with oxides of lead and tin, the spectrum lines of the lead and tin serving as internal standards with which lines due to the constituents of the ash were compared. By comparison of the spectra obtained with those of standard synthetic mixtures (*i.e.* synthetic ash plus lead and tin oxides) rough estimates of the composition of a fair number of ashes—Empire and non-Empire, lamina and stalk, in all about 80 samples—were made. From this examination it appeared that certain chemical differences existed between different types of tobacco ash. As these differences appeared to be characteristic and not random, a detailed examination was made of a larger number of samples, as follows:

Light Virginian type from source A, lamina 26, stalk 23; from source B, lamina 22, stalk 10; from source C, lamina 50, stalk 40; from source D, lamina 13, stalk 13. *Indian type* from source E, lamina 22. *Turkish type* from source F, combined lamina and stalk 10; from source G, lamina 8, stalk 8. *Miscellaneous types* 16. Total 261 samples.

EXPERIMENTAL

Ashing the Tobacco.—The tobacco was burned at a low temperature and the ash was treated with ammonium carbonate, and re-heated. The resultant ash was finely ground. Early analyses showed that occasionally carbonation was incomplete; consequently the practice was adopted, and is followed in this paper, of giving the major metallic constituents of the ash not as percentages of metal, oxide, or carbonate on the ash, but as ratios one to another, *e.g.* Ca/K, Mg/K and Mg/Ca. In this way irregularities due to variations in the nature and amounts of the acid radicles present are avoided.

Determination of Ca, K, Mg, Sr and Mn in the Acid Extract by the Lundegardh Method.—An apparatus similar in its essential features to that described elsewhere⁴ was constructed. The principle of the method is that the material to be examined is dissolved in some convenient liquid and the solution so obtained is atomised in the air supply of an air-acetylene flame. Conditions of the flame, *e.g.* air and acetylene pressures, are standardised and the spectra are photographed with a Hilger Medium Spectrograph, using Zenith plates. Actually 0.1 g. of the ash was treated with 10 ml. of water and 4.5 ml. of 4 *N* hydrochloric acid, allowed to stand on the steam-bath for $\frac{1}{2}$ hr., boiled for 2 mins., diluted and filtered and the filtrate was made up to 100 ml. This solution gave spectra of suitable density for potassium, magnesium, strontium, and manganese determinations. For the determination of calcium a (1+10) dilution was made. Duplicate spectra (1 min. exposures) of 10 samples and 6 standard solutions were photographed on one plate. The blackening of the spectrum lines (due to Ca, K, Mg, Sr and Mn) and that of the spectrum background in the vicinity of the lines were measured, using, in the earlier part of the work, a microphotometer previously described,⁵ and later a Hilger microphotometer. The measurements so obtained were treated in the usual way, *i.e.* the ratio:

deflection for line / deflection for background

was plotted against percentages of the element present in the six standards, and from the curve drawn through the seven points (six from the standards, the seventh point corresponding to zero percentage and ratio 1.0) the percentages of the element present in the 10 samples were read off. The amounts of calcium, potassium and magnesium were determined throughout in this way. The amounts of strontium and manganese, which were small, were only occasionally determined by measurement; usually the plate was examined visually and the percentages of these two elements were estimated. The errors of the Lundegardh method are usually less than ± 5 per cent. of the amount of the material determined, their precise magnitude depending upon the element determined, its

environment and its absolute concentration in the solution aspirated into the air stream. With standard solutions for which the preliminary physical and chemical treatment was well controlled, this degree of precision was obtained. The errors of the complete routine procedure, *i.e.* extraction of 0.1 g. of ash, filtration, etc., followed by spraying, were not unexpectedly greater. The repeatability of the whole process was tested by making a number of repeat determinations after some weeks or months. Table I gives typical results:

TABLE I

| Sample No. | K Per Cent. | Ca Per Cent. | Mg Per Cent. | Ca/K | Mg/K | Mg/Ca |
|------------|----------------|-----------------|-----------------|------|------|-------|
| 62 | 22.3 | 10.2 | 3.5 | 0.46 | 0.16 | 0.34 |
| | 22.0 | 11.5 | 3.5 | 0.52 | 0.16 | 0.30 |
| 90 | 15.6 | 13.5 | 3.0 | 0.87 | 0.19 | 0.22 |
| | 18.5 | 15.0 | 3.0 | 0.81 | 0.16 | 0.20 |
| 160 | 15.2 | 18.5 | 3.5 | 1.22 | 0.23 | 0.19 |
| | 18.0 | 21.0 | 3.5 | 1.17 | 0.19 | 0.17 |
| 226 | 8.2 | 14.0 | 3.0 | 1.71 | 0.37 | 0.21 |
| | 8.8 | 15.9 | 3.0 | 1.89 | 0.34 | 0.19 |

Estimation of Na, Li, Al, Si, B, Pb and Ba by the Arc Method.—About 10 mg. of ash were placed on the lower, slightly hollowed cathode of a pair of copper electrodes and the arc (200 v.; 2 amps.; gap 4 mm.) was struck. Duplicate exposures, each of 2 mins., were photographed, with Ilford Special Rapid Panchromatic plates, the spectrograms obtained were compared visually with those of synthetic mixtures, and estimates of the amounts of the elements present were made. To avoid the implication of a degree of accuracy which does not exist, we have preferred not to give the amounts so estimated as percentages, but to distinguish, for each element, only four (or three) grades of quantity, designated by the bracketed numerals (1), (2), (3) and (4). The approximate percentages to which they correspond, for the various elements, are shown in Table II.

TABLE II

| | Na | Li | Al | Si | B | Pb | Ba | Sr* | Mn* |
|-----|------|--------|------|----|-------|-------|--------|------|-------|
| (1) | <0.1 | <0.005 | <0.5 | <5 | <0.02 | <0.03 | <0.005 | <0.1 | <0.03 |
| (2) | 0.25 | 0.005 | 0.5 | 8 | 0.02 | 0.03 | 0.03 | 0.3 | 0.1 |
| (3) | 0.5 | 0.01 | 2.0 | 12 | 0.08 | 0.15 | 0.14 | 0.8 | 0.2 |
| (4) | — | 0.04 | 4.0 | 16 | 0.40 | 0.8 | | | |

* Sr and Mn were visually estimated from Lundegårdh plates.

Results of high precision were not obtainable from this simple arc, but duplicates taken at different times on different plates never differed by more than one grade, *e.g.* (2) repeated as (3), or (4) repeated as (3), and usually differences were of nil or a half grade, *e.g.* (2) repeated as (2), or (3) repeated as (3)–(4). More precise procedures, involving the addition to the ash of a standard amount of some element which gives lines to serve as internal standards, do of course exist,* but for this exploratory work the gain in accuracy did not appear to be worth the additional labour and loss of speed that quantitative arc methods involve. It must be remembered that the probable errors of derived averages, distributions, and such values are very much less than the errors of the individual determinations.

To avoid systematic errors the samples were analysed in irregular order over a long period of time, and by three operators, one working over the whole period and two part time.

* See, for example, "Spectrographic Analysis in Great Britain," A. C. Candler (Adam Hilger, Ltd.), p. 47; "Paints, Varnishes and Fabrics," by War Department Chemist. This method is used by one of us for the general analysis of non-conducting powders.

RESULTS

The *average* composition of the major groups of tobacco ashes examined are summarised in Table III.

TABLE III

| Origin | No. of samples examined | Ca/K | Mg/K | Mg/Ca | Li | Al | B | Si | Mn | Na | Sr | Pb | Ba |
|--------------------|-------------------------|------|------|-------|-----|---------|---------|---------|---------|---------|---------|---------|---------|
| A Lamina | 26 | 0.90 | 0.15 | 0.16 | (3) | (2)-(3) | (2) | (2) | (1) | (1)-(2) | (1)-(2) | (1) | (1)-(2) |
| B " | 22 | 1.50 | 0.35 | 0.25 | (2) | (2) | (2) | (2) | (1) | (1) | (2) | (1) | (1)-(2) |
| C " | 50 | 1.58 | 0.23 | 0.15 | (2) | (2)-(3) | (2)-(3) | (3) | (2) | (1)-(2) | (1) | (3)-(4) | (1) |
| D " | 13 | 1.96 | 0.20 | 0.10 | (1) | (2) | (2)-(3) | (1) | (2) | (1)-(2) | (1) | (3)-(4) | (1) |
| E " | 22 | 2.88 | 0.58 | 0.20 | (4) | (2) | (2)-(3) | (2) | (1) | (1)-(2) | (2) | (1) | (2) |
| F Lamina and stalk | 10 | 3.64 | 0.39 | 0.11 | (4) | (4) | (2)-(3) | (2)-(3) | (1)-(2) | (2) | (1)-(2) | (1) | (1) |
| G Lamina | 8 | 1.72 | 0.49 | 0.33 | (1) | (2)-(3) | (2)-(3) | (4) | (2) | (1)-(2) | (1)-(2) | (1) | (1) |
| A Stalk | 23 | 0.36 | 0.11 | 0.33 | (3) | (2) | (2) | (1) | (1) | (1)-(2) | (1)-(2) | (1) | (2) |
| B " | 10 | 0.61 | 0.17 | 0.28 | (2) | (2) | (2) | (1) | (1) | (1) | (2) | (1) | (2)-(3) |
| C " | 40 | 0.58 | 0.12 | 0.21 | (1) | (2) | (2) | (1) | (1) | (1)-(2) | (1) | (1) | (1)-(2) |
| D " | 13 | 0.89 | 0.17 | 0.19 | (1) | (2) | (2) | (1) | (1) | (1) | (1) | (1)-(2) | (1)-(2) |
| G " | 8 | 0.96 | 0.36 | 0.39 | (1) | (2) | (2) | (2) | (1) | (1) | (2) | (1) | (1) |

Strong evidence that the differences revealed between the various ashes are real and not fortuitous is obtained from the "half-means," *i.e.* the averages of the first half and of the second half of the total number of samples examined. Table IV gives a number of these half-means for the ratio Ca/K.

TABLE IV

| Tobacco | No. of samples | First half-mean | Second half-mean | Final average |
|-----------|----------------|-----------------|------------------|---------------|
| C Lamina | 50 | 1.68 | 1.47 | 1.58 |
| C Stalk | 40 | 0.57 | 0.59 | 0.58 |
| H* Lamina | 44 | 1.03 | 1.22 | 1.13 |
| H* Stalk | 35 | 0.39 | 0.45 | 0.42 |

* Includes A, B, and three miscellaneous samples.

It is interesting that though the values of Ca/K for the different types of lamina and stalk differ so markedly, the average value of the ratio, *R*, of Ca/K in stalk to Ca/K in lamina, for the five groups of tobacco ashes for which data are available, is relatively constant, as shown by Table V.

TABLE V

| Tobacco | Number (<i>n</i>) of samples of (lamina and stalk) | \bar{R}/n |
|------------------------|--|-------------------------|
| A Light Virginian Type | .. 23 | 0.45 |
| B " " " | .. 10 | 0.41 |
| C " " " | .. 40 | 0.43 |
| | | (Half-means 0.40, 0.45) |
| D " " " | .. 13 | 0.45 |
| G Turkish Type | 8 | 0.62 |

These findings, taken in conjunction with Table III, reveal that for light Virginian-type tobacco: (1) the ash of the lamina and of the stalk show marked differences of average composition dependent upon the area of origin, *i.e.* upon *external* conditions such as soil, climate and cultivation; (2) though the average compositions of lamina and of stalk from different places differ, the average value of the ratio of Ca/K in stalk to Ca/K in lamina, which is governed by the *internal* balance and economy of the plant, appears to be substantially independent of external conditions, and approximately equal to 0.43.

IDENTIFICATION OF TOBACCO

PERCENTAGE DISTRIBUTION TABLES.—Table III shows that certain distinct differences exist between tobaccos from various areas, but *average* values would be of diagnostic use in only a small percentage of the cases encountered. To attempt identification or discrimination, the construction of distribution tables is essential. This has been done for A, B, C, and E laminae, and for A and C stalks, since data were derived from comparatively large numbers (22–50) of samples, and it is maintained that these may be used in processes of identification. Tables VI and VII, which are self explanatory, are typical percentage distribution tables.

TABLE VI. DISTRIBUTION TABLE FOR Ca/K IN STALK

| | Percentage of samples examined for which Ca/K falls within range: | | | | | | | |
|--------|---|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|--------------------|
| | 0 to 0.20 | 0.21 to 0.40 | 0.41 to 0.60 | 0.61 to 0.80 | 0.81 to 1.00 | 1.01 to 1.20 | 1.21 to 1.40 | 1.41 to 1.60 |
| Source | | | | | | | | |
| A | 4 | 61 | 26 | 9 | 0 | 0 | 0 | 0 |
| C | 2 | 23 | 23 | 33 | 10 | 7 | 0 | 2 |

TABLE VII. DISTRIBUTION TABLE FOR SR

| Source | Lamina or stalk | Percentage of samples with Sr contents: | | |
|--------|--------------------|---|-----|-----|
| | | (1) | (2) | (3) |
| A | L | 73 | 19 | 8 |
| B | L | 23 | 61 | 16 |
| C | L | 90 | 10 | 0 |
| E | L | 31 | 57 | 11 |
| A | S | 65 | 30 | 5 |
| C | S | 95 | 3 | 2 |

Diagnostic Use of Distribution Tables.—The diagnostic value of data of this type clearly depends upon whether the correlation and interdependence of the different characteristics are high or low. In the former circumstances the diagnostic value would be low; in the latter high. For example, the question may be put:—If a lamina ash from source C were to have a low Ca/K value which would tend to put it in the A group, would it also be likely to have other A characteristics (such as high Sr, Ba, Li, and low Pb, Mn, Si) or would the other properties be unassociated with the Ca/K value, so that a fair probability would exist of their values restoring the balance and bringing the sample back into its true group? No doubt this question could be answered by a statistical search for, and analysis of, the degree of correlation between the various characteristics. The alternative method, an outline of which is given below, is to attempt to determine the country of origin of a tobacco by means of the chemical analysis of the ash and the distribution tables given.

Ten ashes of tobacco from source A (both of lamina and of stalk), 10 from source C (both of lamina and of stalk) and 10 from source B (lamina only) were chosen as fairly as possible from the numbers previously examined; for example, every fourth sample of C source tobacco was chosen, and the B source laminae selected were those for which, by chance, the stalk ash had also been analysed. The problem set was to allot each of the 30 laminae to one of the three groups A, B, or C, and each of the 20 stalks into either the A or C group. This test appeared to be a fair one, since it was the sort of problem likely to be met in practice, and the characteristics of the 3 groups (see Table III) are not so widely different as those of some groups which might have been chosen, *e.g.* A and E.

From the distribution tables, probability factors may be derived. Consider, for example, strontium in the lamina ash. From Table VI we have

| Source | Percentage with Sr contents: | | | Totals |
|--------|------------------------------|-----|-----|--------|
| | (1) | (2) | (3) | |
| A | 73 | 19 | 8 | 100 |
| B | 23 | 61 | 16 | 100 |
| C | 90 | 10 | 0 | 100 |
| Totals | 186 | 90 | 24 | — |

Hence probability factors are:

| Source | Sr (1) | Sr (2) | Sr (3) |
|--------|-----------------|----------------|----------------|
| A | $73/186 = 0.39$ | $19/90 = 0.21$ | $8/24 = 0.33$ |
| B | $23/186 = 0.12$ | $61/90 = 0.68$ | $16/24 = 0.67$ |
| C | $90/186 = 0.48$ | $10/90 = 0.11$ | — |
| | 0.99 | 1.00 | 1.00 |

Thus if a sample of lamina ash is found to contain strontium of amount (2), this indicates that the sample is most probably from B (probability factor 0.68), less probably from A (factor 0.21), and least probably from C (factor 0.11). Ideally, smooth distribution curves would be drawn from the distribution table data, and ideal, instead of actual, percentages read off from these curves and used for deriving probability factors; but the number of samples examined is not sufficiently large to allow the precise form of the distribution curve to be fairly drawn. Consequently it has been preferred to deduce probability factors from the percentage distributions which were actually determined by the examination of, say, N samples, rather than from percentages estimated for $N \rightarrow \infty$. In all, probability factors were derived in this way for Ca/K, Mg/K, Mg/Ca, Sr, Ba, Pb, Mn, Li and Si for laminas, and for Ca/K, Mg/K, Mg/Ca, Sr, Ba, Li for stalks, these being the characteristics which showed the greatest variations dependent upon area of origin.

The probability factors for each of the 50 samples chosen were assessed and their average values were taken. Below are given two typical examples.

| Sample of lamina from source C | | | | Sample of stalk from source C | | | |
|--------------------------------|------|------|------|-------------------------------|------|------|--|
| Probability factors | | | | Probability factors | | | |
| Analysis | C | B | A | Analysis | C | A | |
| Ca/K 1.32 | 0.23 | 0.58 | 0.19 | Ca/K 0.48 | 0.47 | 0.53 | |
| Mg/K 0.14 | 0.43 | 0.14 | 0.43 | Mg/K 0.04 | 0.49 | 0.51 | |
| Mg/Ca 0.11 | 0.37 | 0.17 | 0.46 | Mg/Ca 0.09 | 1.00 | — | |
| Sr (1) | 0.48 | 0.12 | 0.39 | Sr (1) | 0.58 | 0.52 | |
| Pb (4) | 1.00 | — | — | Ba (1) | 0.61 | 0.39 | |
| Ba (1) | 0.34 | 0.31 | 0.35 | Li (1) | 0.68 | 0.32 | |
| Mn (2) | 0.61 | 0.03 | 0.36 | | | | |
| Li (2) | 0.36 | 0.32 | 0.32 | | | | |
| Si (2) | 0.22 | 0.46 | 0.32 | | | | |
| Average probability factors | 0.45 | 0.24 | 0.31 | Average probability factors | 0.64 | 0.36 | |

It will be observed that in both instances the samples are correctly identified as from source C.

The outcome of applying this method to the 50 samples was:

| Number of samples | Type | Correctly identified |
|-------------------|----------|----------------------|
| 10 | A Lamina | 9 |
| 10 | A Stalk | 8 |
| 10 | C Lamina | 7 |
| 10 | C Stalk | 10 |
| 10 | B Lamina | 9 |
| Totals | 50 | 43 |

TATE AND WHALLEY: THE SPECTROGRAPHIC ANALYSIS OF TOBACCO ASH

An earlier attempt at identification, in which the probability factors for the characteristics Mg/K and Mg/Ca were not included, gave 40 correct out of 50. It appears possible, therefore, that if all the characteristics were to be included the probability of a correct identification being made would increase. Such a comprehensive assessment, using all the data available, was not thought to be worth the additional labour involved, since already the desired principle had been demonstrated, namely, that a method of identification with a high probability of success, of the place of origin of a tobacco, may be based upon the analysis of its ash.

The method would be improved by the use of more precise methods of analysis, particularly in the arc technique, and by the accumulation of more data. As a standard procedure for indicating the source of an unidentified sample it may be necessary to add at intervals to the collected data those for new known samples from current production, at the same time removing from use the data for an equal number of the earliest samples. This would accommodate the method to any gradual changes which may be in progress.

Though 43 out of the 50 samples chosen were correctly identified, it should be remembered that by tossing a coin or spinning a roulette wheel the chances are that the correct answer would have been made in $\frac{30}{3} + \frac{20}{2} = 20$ cases out of the 50 chosen.

SUMMARY.—Analyses have been made of over 300 samples of tobacco ash. New data have been obtained on the inorganic constituents and on their dependence or otherwise upon the area of origin. The *average* composition of the ashes of tobaccos from certain different areas showed distinct differences. From a statistical treatment of the results, probability factors were derived which had high diagnostic value. It is demonstrated that a method of identifying the source of tobacco, with a high probability of success, may be based on analysis of the ash.

We are indebted to Drs. McClelland and Fay who at different times helped with the experimental work, and to Mr. G. H. Dyer for practical assistance throughout.

We thank the Government Chemist, Dr. J. J. Fox, C.B., O.B.E., for his permission to publish this account.

REFERENCES

1. E.g. C. Pyriki, *Z. Unters. Lebensm.*, 1939, **78**, 162; J. Vladescu and N. Dimofte, *id.*, 1936, **71**, 358.
2. E.g. F. R. Darkis, L. F. Dixon, F. A. Wolf and P. M. Gross, *Ind. Eng. Chem.*, 1937, **29**, 1030; C. Coolhaas, *Proefstat Vorstetenland. Tabak Med.*, 1938, **85**, 73.
3. H. K. Whalley, *Trans. Oil and Colour Chemists' Assoc.*, 1939, **22**, No. 223.
4. H. Lundegardh, *Lantsbrukshogskolans Annaler*, 1936, **3**, 49; "Die Quantitative Spektralanalyse der Elemente," Jena, 1929-34, Bd. I-II.
5. H. K. Whalley, *J. Soc. Chem. Ind.*, 1937, **56**, 177r.

September, 1940

The Analytical Constants of Spirit Vinegar

By C. H. MANLEY, M.A., F.I.C.

RECENT experience has indicated that appreciably lower figures must be accepted for the minimum percentages of ash, nitrogen and phosphoric oxide in spirit vinegar than those put forward by Edwards and Nanji¹ in their valuable paper on the oxidation, iodine and ester values of vinegars. Also, under certain conditions, the oxidation value, but not the iodine value, may come within the malt vinegar range.

As a sequel to the mislabelling of bottles of spirit vinegar on sale in the Leeds area as "Genuine Vinegar (Non-Synthetic)," visits were paid to the wholesalers and to the manufacturers' distributing centre, it being considered that a product described as above should be malt vinegar. It was then learnt that the manufacturers were supplying a spirit vinegar concentrate containing approximately 12 per cent. of acetic acid to the wholesalers, who were then diluting it with water to three volumes prior to distribution to local shopkeepers, including fish and chip friers. The manufacturers also, when required, were themselves prepared to supply the diluted article, which was then quite properly labelled "Spirit Vinegar" in bold type.

The wholesalers subsequently agreed to label their product in similar manner as "Genuine Spirit Vinegar" and to omit the words "Non-Synthetic," previously printed in very small type.

The following table gives the percentage (g. per 100 ml.) results of the analysis of various samples obtained direct both from the manufacturers and from the wholesalers. The products supplied by the manufacturers may be accepted as genuine spirit vinegar.

TABLE I

| Source | Acetic acid | Total solids | Ash | Nitrogen | P ₂ O ₅ | Oxid. value | Iodine value |
|---|-------------|--------------|-------------|-------------|-------------------------------|-------------|--------------|
| 1. Manufacturers' concentrate | 11.94 | 0.17 | 0.032 | 0.003 | 0.005 | 464 | 30 |
| 2. Wholesalers' concentrate | 11.80 | 0.55 | 0.044 | 0.016 | 0.005 | 272 | 38 |
| 3. Manufacturers' spirit vinegar | 4.14 | 0.20 | 0.024 | 0.006 | 0.003 | 142 | 21 |
| 4. Wholesalers' spirit vinegar (A) | 3.92 | 0.43 | 0.044 | 0.007 | 0.004 | 1120 | 46 |
| 5. Do. (B) | 3.90 | 0.26 | 0.028 | 0.006 | 0.002 | 211 | 14 |
| 6. Do. (C) | 4.12 | 0.20 | 0.018 | 0.011 | 0.003 | 109 | 13 |
| 7. Do. (D) | 4.27 | 0.19 | 0.016 | 0.005 | 0.003 | 159 | 18 |
| 8. Water from rinsed cask before making (C) | — | — | — | — | — | 3 | 4 |
| Spirit vinegar, normal range | — | 0.16-0.30 | 0.016-0.090 | 0.005-0.040 | 0.002-0.03 | — | — |
| Do. (Edwards and Nanji) | — | 0.16-0.30 | 0.04-0.09 | 0.03-0.04 | 0.002-0.03 | 88-225 | 8-27 |
| Artificial vinegar, normal range | — | 0.1-0.5 | 0.005-0.05 | nil-0.04 | nil-0.03 | 1-16 | 2-252 |
| Do. (Edwards and Nanji) | — | 0.30-0.45 | 0.02-0.05 | do. | do. | do. | do. |

Apart from the ash of No. 4 (the somewhat exceptional character of which is discussed below), all the samples of spirit vinegar examined had values for ash, nitrogen and phosphoric oxide well below the minima cited by Edwards and Nanji.

But for their oxidation values these vinegars would ordinarily be classified as artificial. The nett effect, therefore, is to render any determinations other than that of the oxidation value of very little use in distinguishing between artificial and spirit vinegars.

It will be seen that, whilst the oxidation value of No. 3 is less than one-third of that of No. 1, the oxidation value of No. 4 is more than four times that of No. 2, the concentrate from which it was prepared, the value in fact being equal to that of a malt vinegar. It transpired that these particular wholesalers make a practice of diluting their concentrate in 60-gallon Australian white wine casks, and that the sample in question represented a mix made in a fresh cask, from the walls of which the spirit vinegar had evidently extracted much of the residual alcohol. Thus pronounced odours of acetaldehyde and iodoform developed during the determinations of the oxidation and iodine values respectively. A sample (No. 5) taken from the second mix in the new cask gave an oxidation value (211) falling within the spirit vinegar range, but actually not much lower than that of the original concentrate. A further fall (to 109) occurred in the third mix (No. 6) made in my presence after the cask had first been washed out by filling with water (No. 8) and emptying. A sample of this water gave very low oxidation and iodine values. On my advice, 20 gallons of this concentrate were mixed with 38 gallons of water instead of with 40 gallons as hitherto, in order that the percentage of acetic acid in the spirit vinegar might not be less than 4 per cent. A further sample (No. 7) bottled off two months later from a subsequent mix made in the same cask showed, contrary to expectation, a rise in the oxidation value to 159, instead of a further fall to a figure between 90 and 100.

Determinations of the iodine value and of the percentage amounts of the chemical constituents, as well as the appearance of the total solids (whether mottled or not) would readily indicate whether a sample with an abnormally high oxidation value were a malt vinegar or otherwise.

The ester values of five of the seven samples of spirit vinegar were determined, and the following results (corrected for a 4 per cent. acid content) were obtained:

TABLE II

| No. | Ester value | Ester value corr. for 4% Acetic acid |
|------------------------------|-------------|--|
| 1 | 24.2 | 8.1 |
| 2 | 17.3 | 5.9 |
| 4 | 3.6 | 3.7 |
| 6 | 5.3 | 5.1 |
| 7 | 4.8 | 4.5 (duplicate sample kept 4 months) |
| Normal figures | 6.0-14.0 | |
| Do. after 18 months' storage | 2.0-3.6 | |

The fixed acidity of No. 4 was only 0.002 per cent. (as tartaric acid), so that it would seem to be impracticable to separate any tartrate extracted from the wine cask. This would apply still more to the three subsequent mixes (Nos. 5, 6 and 7).

It is obvious that the ester values will not afford much help with the problem, although they show a slight decrease after the vinegars have been kept for nine months. The determination of the oxidation value, however, still remains the most sensitive test for distinguishing between spirit and artificial vinegars.

On the other hand, it would appear possible to pass off artificial vinegar as spirit vinegar by diluting coloured strong acetic acid in a wine cask, provided that the operation were not repeated more than once.

REFERENCES

1. F. W. Edwards and H. R. Nanji, *ANALYST*, 1938, **63**, 410.
2. E. T. Illing and E. G. Whittle, *id.*, 1939, **64**, 329.
3. H. Amphlett Williams, *id.*, 1939, **64**, 270.

CITY ANALYST'S LABORATORY
1, SWINEGATE, LEEDS, 1

June, 1940

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

DETECTION OF BEEF FAT IN LARD (BÖMER NUMBER)

A NEW modification of this method, proposed by an American Chemical Society Committee, has recently appeared (Abst., ANALYST, 1940, 65, 508), but the full description is not sufficiently detailed to ensure satisfactory deductions from the results it yields.

Attention may be drawn, first, to the following point. It is directed that 20 g. of lard are to be treated with acetone at 30° C. and the solution diluted to 100 ml. with acetone at 30° C. It is evidently intended that solution should be complete and the liquid clear at this point.

The solution is then kept at 30° C. for 18 hours. Crystals are frequently deposited during this period, but I have encountered a large number of samples in which no crystals formed, and in some of these the lard was known to contain beef fat in admixture. Since absence of crystals is taken at present to indicate absence of hard fat, it follows that some adulterated lards may be classed as pure with the test in its present form. The difficulty originates in the abnormal stability of some supercooled lard solutions, and may be simply overcome by making a clear solution of the lard as directed, cooling it until crystallisation has started, and then placing it in the constant-temperature bath at 30° C. for 18 hours. Variations in the speed with which the initial crystallisation is allowed to take place do not affect the m.p. of the glycerides or fatty acids, and it makes no difference whether cooling is allowed to proceed until a heavy deposit has formed or is checked while the deposit is still small. Similar difficulties arise with regard to the induction of crystallisation in the corresponding B.P. test in which ether is used as solvent.

Secondly, a temperature variation of 4 degrees is allowed in both the A.C.S. method and the B.P. method during the standing period. Crystals may often be obtained at the lowest permitted temperature but not at the highest. If such crystals are of low melting-point, as may happen, conflicting deductions can be drawn from the results of either method. Such conflict would be avoided if temperature variations permissible were reduced to a fraction of a degree. It is a simple matter to maintain the fat solution within $\pm 0^{\circ}\text{C.}$ of a mean temperature in a constant-temperature bath, and mean temperatures of 30° C. (A.C.S. method) and 18° C. (B.P. method) are suggested as most satisfactory.

K. A. WILLIAMS

6, MILNER STREET
LONDON, S.W.3

October 9th, 1940

SAMPLING OF PRINTED PAPER

ON several occasions, in circumstances in which sampling has been difficult, these laboratories have had to determine poisonous metals in a sheet of printed paper to be used for wrapping foodstuffs. Unless the printed design is symmetrical, the sheet cannot be sampled by cutting out a portion of it, for there may be as many as eight or ten printings, each with a different ink, any of which may be responsible for the poisonous metal-content of the sheet. Destroying the whole of such a sheet by wet oxidation and taking an aliquot part of the resulting solution is one way out of the difficulty, but wet oxidation is inconvenient if the sheet is large and, further, if the whole sheet is used at once, there is no opportunity for replicate determinations unless more sheets are available. Even then, one cannot be certain that all sheets will carry the same amount of ink, as the thickness of the ink film printed may vary during a printing run. The need of some method of dividing a single sheet of printed matter to obtain a representative sample will therefore be apparent. A method of sampling which has been found to give satisfactory results is as follows:

The whole of the sheet is cut up into small pieces, about 2 or 3 mm. square. This is the most tedious part of the operation and is best carried out by first cutting the sheet into strips on a guillotine and then cutting the strips across. The pieces are then mixed by an air blast in a Buchner flask fitted with a cork and a lead-in tube for the air reaching to the bottom of the flask. About two or three minutes' mixing, while the flask is shaken at the same time to break up drifts, will generally be sufficient. Obviously care must be taken not to have the air blast so strong that the paper is blown out of the side tube of the flask and, in addition, the air should not be either very dry or very wet, otherwise the moisture-content of the paper may be altered. After mixing, the paper pieces can be turned out of the flask and quartered in the usual manner until a sample of convenient size is obtained.

The method was tested by taking known weights of four differently coloured papers, cutting them up, mixing in the manner described, quartering, and analysing the sample thus obtained by hand-picking to determine the proportion of each colour present. The results obtained showed a sampling error of about ± 6 per cent, the extremes being $+6.6$ per cent. and -6.5 per cent. A typical series of results is as follows:

Substance of paper, 52 g. per sq. metre; size of pieces, 2 to 3 mm. square; number of quarterings, 2.

| Paper | Wt. taken g. | Wt. found in quartered sample g. | Per cent. wt. found | Deviation from mean Per Cent. |
|-------------|-----------------|--|---------------------|-------------------------------------|
| | | | Per cent. wt. taken | |
| Blue | 0.629 | 0.163 | 25.9 | +4.0 |
| Orange .. . | 0.613 | 0.144 | 23.5 | -5.6 |
| Yellow .. . | 0.508 | 0.123 | 24.2 | -2.8 |
| Pink .. . | 0.609 | 0.158 | 25.9 | +4.0 |
| Total .. . | 2.359 | 0.588 | Mean 24.9 | |

The error involved is probably larger than the experimental error in some analytical determinations which would be carried out on the sample thus obtained, but for many purposes, *e.g.* determining whether the sheet complies with a specified limit for lead or arsenic, the accuracy is sufficient.

An attempt to reduce the sampling error by cutting the sheet into smaller pieces, 1 mm. square, gave no improvement in accuracy and the cutting up process then became impossibly tedious for any ordinary purpose. This drawback might be overcome by grinding the paper if a suitable machine were available. The method was tested on light-weight papers (52 g. per sq.m.) and heavier papers (460 g. per sq.m.), and a similar degree of accuracy was obtained in every instance.

I wish to thank the Council of the Printing and Allied Trades Research Association for permission to publish these results.

J. H. YOUNG

PATRA HOUSE

10, ROBIN HOOD COURT

SHOE LANE, LONDON, E.C.4

August, 1940

Notes from the Reports of Public Analysts

The Editor would be glad to receive the Annual or other Reports of Public Analysts containing matter of special interest to the Society. Notes made from such Reports are submitted to the Publication Committee.

CITY OF BIRMINGHAM

ANNUAL REPORT OF THE CITY ANALYST FOR THE YEAR 1939

"CHEESE SPREADS."—Three samples bore an unsatisfactory label on which the fat contents were stated to be "25 per cent. i.d.s." as a minimum. The three letters were intended to mean "in dry solids," but the ordinary purchaser would be apt to imagine that the actual amount of fat present was 25 per cent., whereas only 12.7, 14.0 and 15.5 per cent. respectively were present, and in each instance the proportion of water exceeded 60 per cent. If it was intended that the amount of fat should be calculated as percentages of the dry solids of the cheese, this should have been stated on the label in plain terms and not in unintelligible abbreviations.

RASPBERRY JAM.—A sample of lower fruit standard raspberry jam was reported as deficient of 34 per cent. of fruit. The manufacturers contended that the correct amount (20 per cent. of the total bulk) was present, and they accounted for the low insoluble solids in the jam by alleging that the variety of raspberries used was liable at times to give an abnormally low figure for these constituents. Such abnormality, however, has only been detected in fruit grown in a particular district in Scotland and picked in late summer.

SODAMINT TABLETS.—Several samples did not agree in composition with formulae in the B.P. Codex. Three samples contained only a trace of ammonium bicarbonate and two contained none at all. The vendors of these were cautioned.

FORMALIN TABLETS.—The B.P. Codex requires each tablet to contain 9.7 mg. of para-formaldehyde. Whilst some samples complied with this formula, several others did not. One sample consisted of three varieties of tablets containing from 0 to 4 mg., of para-formaldehyde, another contained only 4 mg., and a third only 2 mg. The vendors were cautioned. As the names sodamint and formalin tablets are given as synonyms for the official Latin names in the Codex, it would prevent confusion if manufacturers would adhere strictly to the formulae and if other names were given to tablets not made according to the Codex prescription. It is unfortunate, to say the least, that a purchaser may get any one of several different articles all sold under the same description.

H. H. BAGNALL

Department of Scientific and Industrial Research

REPORT OF THE BUILDING RESEARCH BOARD FOR THE YEAR 1939*

DESCRIBING the functions of the Research Station in war time the Chairman of the Board (Mr. G. Mowlem Burt) mentions that it is (1) supplying data relative to a particular problem, (2) investigating new materials and methods of construction, and (3) investigating the use of alternative materials. The Director (Dr. Stradling) was asked during the year to undertake the duties of Chief Adviser on Research and Experiment to the A.R.P. Department.

In his Report on General Research and Investigation the Director gives an outline of further work on various problems.

CLEANING OF BUILDINGS.—The method of continuous spraying has been successfully applied to more buildings, including parts of Trinity College and other Cambridge colleges. It has also been successfully used on concrete buildings in London. The stains that developed on the Admiralty Screen, Whitehall, after it had been cleaned have faded appreciably during the year.

EFFECT OF LIME IN FIRED CLAY PRODUCTS.—The defect known as "lime-blowing" is caused by the expansion of nodules of lime or calcium sulphate as they become hydrated. It is usually prevented by grinding the clay very finely or by "docking," i.e. soaking the bricks or tiles in water when they are taken from the kiln. Local conditions must determine which method is the more suitable. In one works, "lime-blowing" could not be induced even by prolonged steaming of bricks after the method of fine grinding had been adopted.

SAND-LIME BRICKS.—It has been found that the properties of the bricks are considerably affected by the state of the silicate bonding material formed during the autoclave process. The silicate may be either wholly crystalline or entirely amorphous. A study of the factors influencing crystallisation should make it possible to control more closely the properties of those bricks.

PROPERTIES OF POROUS BODIES.—This investigation has proceeded along three main lines:

(1) *Crystallisation of Salts within the Pores.*—The "crystalline test" (in which the material is immersed in sodium sulphate solution and then dried to induce crystallisation in the pores, the process being repeated several times) has enabled the weathering qualities of natural building stones to be assessed and has provided a means of distinguishing good limestones or bricks from those of inferior quality.

(a) *Effect of the Temperature of the Sodium Sulphate Solution.*—It has been found that at temperatures above 30° C. no disintegration of natural stones occurs in the crystallisation test. This is due to the fact that 30° C. is higher than the transition temperature of sodium sulphate decahydrate, the formation of which within the pores can set up stresses greater than the tensile strength of natural stones.

(b) *Rate of Accumulation of Salt in the Pores.*—When samples of the stones were tested with the sodium sulphate solution above 30° C. there was for each specimen an initial increase in the salt content, but finally a limiting constant value was reached.

(c) *Diffusion of Salt from the Stone.*—When a specimen of stone containing salt was placed in water, the rate of diffusion of the salt varied not only with its concentration, but also with the type of stone.

(2) *Relation between Frost-resistance and Pore Structure.*—An apparatus has been devised for measuring the changes in linear dimensions of a material when the crystals are formed within the pores. It has been found that the formation of ice within the pores of clay bricks results in an increase in linear dimensions. The nature of the change varies with the type of brick.

ARTIFICIAL WEATHERING OF ASPHALT.—By removing the normal surface finish obtained by sanding the ultimate degree of whiteness was more rapidly obtained. The removal of the surface was effected by cutting and grinding with carborundum.

FINESS OF PORTLAND CEMENTS.—An air permeability method for measuring the surface area of cements has been found to afford a rapid means of estimating the specific surface of fine powders (*cf.* Lea and Nurse, *J. Soc. Chem. Ind.*, 1939, 58, 278).

RESISTANCE OF CONCRETE AND MORTAR TO ATTACK BY CHEMICAL AGENTS.—Waste waters from the milk industry were found to cause softening of Portland cement mortars, whereas high alumina cement was entirely unaffected (Lea and Bessey, *Concr. Constr. Eng.*, 1939, 34, 610).

"CRAZING" OF CAST CONCRETE PRODUCTS.—A quick laboratory method of testing the liability of cast concrete to "craze" has been devised, the samples being alternately wetted and dried in a cycle of treatments under standard conditions; it has given promising results.

UTILISATION OF BLAST FURNACE SLAG.—Slag has been used for many purposes as an aggregate of concrete. The only trouble so far reported appears to be due to the presence of small amounts of sulphate producing expansion in the concrete. A maximum of 0.5 per cent. of sulphate, expressed as SO_3 , in the aggregate of sand size is apparently a safe permissible limit.

PATTERN-STAINING OF CEILINGS.—Aitken's investigations (*Proc. Roy. Soc. Edinb.*, 1884, 12, 440) proved that dust tends to move from hot surfaces and to attach itself to cold ones. They

* H.M. Stationery Office, York House, Kingsway, London, W.C.2. Price 1s. net.

afford a satisfactory explanation of the stains on plaster ceilings, in which the pattern of the construction behind the plaster is revealed. Experiments described dispose of the fallacy that the patterns are due to dust passing through the plaster. Recently there have been numerous instances of pattern-staining on hollow-tile ceilings. To prevent this, sufficient thermal insulation must be provided to reduce the heat flow through them to the level of the flow through the hollow tiles.

THE TESTING OF AIR FILTERS.—Two sets of apparatus for testing the efficiency of air filters have been devised. One of these is portable and is primarily designed for filters in commercial use; the other is a laboratory plant capable of accommodating single filter-units, which are usually 20 in. square.

New South Wales

ANNUAL REPORT OF THE GOVERNMENT ANALYST FOR THE YEAR 1937

THE Government Analyst (Mr. S. G. Walton) reports that 34,569 samples were examined under the Pure Food Act and 2292 for the Public Services. The food samples consisted principally of milk (25,903), and of these, 369 were adulterated. Among the points of interest discussed in the Report are the following:

BREAD STANDARDISATION.—Representations were made by an Inter-State Nutritional Advisory Board, to investigate the possibility of producing an absolutely standard white flour for bread. It was found that in New South Wales the nitrogenous content of wheat materially increased from the south to the north; hence the production of an absolutely uniform bread appeared impracticable, although the amended standards for wheatmeal, wholemeal and brown bread provide for products of approximately uniform composition. It might be advisable, however, to increase the prescribed minimum nitrogen-content of flour from the present figure of 1.2 per cent.

SULPHUR DIOXIDE IN BEER.—The Regulations allow preservative to be added to bulk beer (if declared), but not to bottled beer or stout. Since traces of sulphur dioxide will generally be found in beers, it is recommended that the presence of total sulphur dioxide in the proportion of not more than 1 grain per gallon should be permitted without declaration; when the amount exceeds that proportion, its presence should be declared on the container.

VEHICLE FOR ESSENCES.—The Regulation permits the use of ethyl alcohol or some other harmless vehicle for essences. A firm asked that consideration should be given to the use of propylene glycol for this purpose, and sent an account of experiments that had been made. It was found, however, that this work had been restricted to one isomer only ($\text{CH}_3\text{CHOH.CH}_2\text{OH}$), and was chiefly concerned with the effect on rats of drinking considerable amounts of that liquid. Propylene glycol is not recognised by the B.P. or B.P.C. for pharmaceutical purposes, and it was decided that, unless there was evidence that it is not more toxic than ethyl alcohol, its use could not be permitted.

GELATINOUS PROTECTIVE COATING FOR FOOD STUFFS.—An application was made for permission to use a gelatin compound hardened with formalin as a protective coating for hams, etc.; it was claimed that the formaldehyde did not penetrate into the meat, and, as the coating would be removed before cooking or eating, no harm could result to the consumers. On investigation it was found that the formaldehyde did penetrate into the meat, and the application was refused. Subsequently an application was received from another firm who wished to use a coating of gelatin hardened with potassium dichromate for the same purpose. In view of the previous experiments, it seemed probable that small amounts of chromium would penetrate into the meat, and, since the addition of chromium compounds to foodstuffs is prohibited under the Pure Food Act, this application was also refused.

ADDITION OF STABILISER TO MILK BEVERAGES.—Application was made for permission to add a stabiliser to chocolate or malted milk sold in bottled form. The stabiliser suggested (sodium alginate) may be regarded as harmless, but the danger of permitting its use is in the creation of a precedent for using this and other substances (gum, gelatin, viscogen, etc.) in cordials, creams, etc., in which its presence may mislead the purchaser into thinking that the thicker the article the better the quality. It was recommended that this aspect of the question should be taken into consideration, and that if the application were granted, a standard should be framed prescribing a minimum milk-content, since such a beverage would enter into competition with fresh milk shakes, etc.

MOULD-CONTENT OF TOMATO PRODUCTS.—A Regulation has been made providing that the mould-content of tomato sauce or purée shall not exceed 50 per cent. of the microscopic fields examined, as determined by the method of the Association of Official Agricultural Chemists, U.S.A. It was proved that the mould-content of tomato pulp depends not so much on the variety of tomatoes as on the efficiency of sorting and trimming.

CARBOXIDE AS A FUMIGANT FOR FOODSTUFFS.—Carboxide is a mixture of ethylene oxide (about 1 part) with carbon dioxide (about 8 parts). The U.S.A. Dept. of Agriculture found that ethylene oxide was highly toxic to insects infesting food products, and no deleterious effects were

noticeable on foods such as dried fruits after exposure for 24 hours to the fumigant. Later it was found that a mixture of 1 vol. of ethylene oxide with 7.15 vols. of carbon dioxide was normally non-inflammable and materially increased the toxicity of the fumigant to insect life. It would appear from these experiments that carboside may safely be used for exterminating insect pests in stored foods.

CAMPHORATED OIL.—Two samples were condemned—one because it had been prepared with maize oil and the other because soya bean oil had been used.

PERCAINE AND PROCAINE.—A medical practitioner prescribed a 0.2 per cent. solution of novocaine in normal saline as a local anaesthetic during an operation. During the operation the patient died, and, as his general condition did not appear to explain the collapse, a death certificate was refused. It was found after the post-mortem and chemical examinations that percaine was present in the organs and that percaine had been substituted for novocaine by the dispenser in making up the prescription. At the Coroner's inquest the evidence showed that the dispenser was under the impression that percaine and novocaine (procaine) are synonymous, and that he was not aware that the former is ten times as toxic as the latter. Unfortunately there are a number of drugs with closely similar names or synonyms, the toxicity of which varies widely. While this is so, accidents of this nature are likely to recur.

Another death from this drug occurred through a patient swallowing some of the percaine used as a local anaesthetic during an operation on the throat.

PASTEURISATION OF CREAM BY THE "FLASH" PROCESS.—As it was suspected that certain cream was not being heated to the stipulated temperature of 178° F. in accordance with the requirements of the Pure Food Act, an investigation was made to ascertain if the chemical tests applied could be relied upon for Court purposes as giving accurate results with regard to the heat treatment of cream. In the experiments, described in detail, the Storch peroxidase test and Kay and Graham's phosphatase test were used. The samples of milk and cream were collected in duplicate, so that a series of tests could be made as soon as possible after collection, and then another series of tests made on the following day, the samples meanwhile being kept on ice. So far as could be ascertained, no alteration took place in the enzymes of the cream, and therefore examination 24 hours after the samples have been collected and kept under proper conditions may be regarded as a reliable guide to the presence, or otherwise, of the enzymes in question. The experiments indicated that the cream under examination had been inadequately pasteurised. No subsequent trouble was experienced, the samples examined conforming with the results of the investigation.

DETERMINATION OF ALCOHOL IN PRESENCE OF ACETONE IN DISTILLATES FROM BLOOD OR URINE.—A method devised by the Government Analyst and R. G. O'Brien has been modified for use in presence of acetone. Denigès found (*J. Pharm. Chim.*, 1899, 9, 7; *Abst.*, ANALYST, 1899, 24, 92) that when acetone is boiled with mercuric sulphate and sulphuric acid it forms a crystalline complex, and in the authors' experience this test is sufficiently accurate to detect and determine less than 1 mg. of acetone in presence of considerable quantities of alcohol. A suitable quantity of the distillate (obtained with a simple apparatus previously described), ranging from 1 ml. to 10 ml. according to the alcoholic content, is diluted to 40 ml. with water, and added to a cooled mixture of 15 ml. of *N*/5 potassium dichromate solution and 20 ml. of conc. sulphuric acid (sp.gr. 1.83) contained in a 100-ml. acetylation flask (fitted with a ground-glass condensation tube, 1 m. long and approx. 1 cm. in diameter). The flask is immersed in water at 78° to 80° C. for 30 minutes, after which its contents are transferred to a 600-ml. flask, the acetylation flask being rinsed with 2 portions of water (100 ml. each) and then with 25 ml. of water, and the washings added to the main solution. The flask is rapidly cooled to room temperature, and its contents are made up to about 500 ml. About 1 g. of potassium iodide is added, and the liberated iodine is titrated with *N*/10 sodium thiosulphate solution. A blank determination is made to obtain the corrected amount (x) of thiosulphate consumed. The mg. of alcohol in the distillate used = x ml. of thiosulphate \times 1.15 - y , where y represents the correction to be applied in presence of acetone as determined by the Denigès method. For each mg. of acetone present 0.029 mg. of absolute alcohol is deducted from the amount found. It is unlikely that in this method a correction of more than 1 mg. of alcohol will ever have to be made.

United Provinces and Central Provinces, India

ANNUAL REPORT OF THE CHEMICAL EXAMINER FOR 1939

THIS is the 75th Annual Report of the Department, which is under the control of Dr. S. N. Chakravarti, F.I.C. During the year under review 3481 cases were investigated as compared with 3185 in 1938. In the medico-legal section 1615 cases were dealt with as against 1524 in 1938.

HUMAN POISONING.—The total number of cases examined was 413, and poison was detected in 60.5 per cent. Of the detected poisons, datura headed the list (37.5 per cent.), followed by opium (23.5 per cent.), arsenic (18.6 per cent.), mercury salts (3.7 per cent.), aconite (3.0 per cent.), cyanides (2.3 per cent.), nux vomica (1.9 per cent.), and alcohol (1.1 per cent.). Other organic poisons found included *Abrus precatorius*, *Lachitra*, yellow oleander, pink oleander and two unidentified vegetable poisons, and among the inorganic poisons were sulphuric and nitric acids and copper sulphate.

ANIMAL POISONING.—There were 36 cases as compared with 23 for the preceding year, and poison was detected in 66.6 per cent. Arsenic was detected in 14 of these cases and kaner (yellow oleander) in 2. There was one unidentified poison.

CREMATION REMAINS.—In cases of alleged poisoning the bones and ashes of five persons were examined, and arsenic was found in four of them.

Government of Madras

ANNUAL REPORT OF THE GOVERNMENT ANALYST TO SEPTEMBER 30TH, 1939

THE Government Analyst (Mr. H. Hawley, M.Sc., F.I.C.) reports that 8833 samples were taken under the Prevention of Adulteration Act (including 1326 examined by the Madras City Analyst). These comprised 2310 of ghee (837 adulterated), 3834 of milk (1878 adulterated), 334 of butter (87 adulterated), 1643 of gingelly oil (237 adulterated), and 712 of other foods (127 adulterated). The total percentage of adulterated samples was 35.8.

ADULTERATION IN MADRAS.—Adulteration in the Province continues to be practised on the grand scale. In some of the panchayats to which the Act has recently been extended more adulterated than genuine food is sold (e.g. in Madanapalle, 75 per cent. adulterated; in Mettupalaiyam, 73 per cent. adulterated). The fact that there is little or no improvement in those areas where the Act has been in force for a number of years appears to be due to the unwillingness of magistrates to impose really deterrent fines. The average fine for the whole Province is only Rs.27, and there are only two areas where it exceeds Rs.50. A fine of Rs.50 at infrequent intervals will not deter even the meanest hawker from foregoing the steady profits that he makes by selling food heavily adulterated with water or a cheap substitute for the genuine article. In General Order No. 346, Public Health, January 29th, 1938, the attention of magistrates was called to the large profits made by food adulteration and to the need for deterrent fines; but the Order appears to have had little or no effect. Another very disturbing fact is that in 9 cases, in which moderate fines of Rs.100 or less have been imposed by the magistrates, these have been reduced on appeal. If the imposition of heavy fines on small vendors is impracticable, it would seem that the Act should provide for imprisonment without the option of a fine when the adulteration is heavy and deliberate.

FREEZING-POINT OF MILK.—The Act prescribes a minimum limit of 9 per cent. of solids-not-fat in buffalo milk, but commonly the figure is 10 per cent., so that such a sample may contain 10 per cent. of added water. All samples showing 10 per cent. or less below the standard figures are examined with the Hortvet cryoscope, and are frequently found to be substantially adulterated.

STEROL ACETATE TEST FOR GHEE.—Different Provinces in India have prescribed standard Reichert values for ghee, ranging from 22 to 30, with which all samples must comply. Actually, genuine ghee commonly gives figures ranging from 20 to 40, though most samples show a more restricted range. In Bombay a minimum Reichert value of 24 is prescribed; all samples giving figures below this are condemned automatically, and the analyst is entitled to pass samples giving a higher figure. In the Madras Laboratory the sterol acetate test, carried out as described in THE ANALYST (1933, 58, 529), is applied to all doubtful samples. Of 376 samples giving Reichert figures between 20 and 24, the test enabled 242 to be classified as genuine and 134 as adulterated, while 70 samples showing figures above 24 were classed as adulterated.

DETENTION OF FOOD HAWKERS.—In 82 cases proceedings against food hawkers had to be abandoned, as the vendors could not be traced. To overcome this difficulty, a provision was inserted in the Public Health Act (Sec. 140) giving an inspector power to detain anyone committing an offence until his identity was satisfactorily established. In practice this provision has been found of little value, because an inspector is unable to tell whether a hawker is selling genuine food or not until he receives the certificate of analysis. This difficulty might be met in

another way. Section 258 of the District Municipalities Act requires that vendors of dairy produce, including hawkers, should be licensed. In most municipalities this section, so far as it applies to hawkers, is a dead letter. If it were operative and hawkers were required to carry a licence, then section 140 of the Public Health Act could be used, as an unlicensed hawker would *ipso facto* be committing an offence and could be held in custody while his identity was being established.

LEAD IN CURRY POWDER.—As the result of English importers insisting on a certificate that curry powder imported into England does not infringe the limit fixed for lead in foods, a number of these powders have been examined. Of 13 samples received from private firms in Madras, 4 contained more than 10 p.p.m., 5 between 5 and 10 p.p.m., and 4 less than 5 p.p.m. A certificate was given when the proportion of lead was less than 5 p.p.m.

DIFFERENTIATION OF HAND-POUNDED FROM MILLED RICE.—It has been prescribed by the Government that hand-pounded rice should be used in hospitals. Of 66 samples received, 35 proved to be milled. When the Order first came into force, the samples examined were almost invariably prepared by milling, whether sold as hand-pounded or not, but owing to adverse reports most samples are now satisfactory.

The following simple method of distinguishing between hand-pounded and milled rice has been worked out by K. V. Sundaram Ayyar, M.Sc., A.I.C. (Senior Assistant to the Government Analyst):—As much rice as will lie on the bottom in one layer, but without completely covering the surface, is put into a 50-ml. beaker. Fifteen ml. of a 5 per cent. aqueous solution of sodium hydroxide are added, and the mixture is stirred by rotating the beaker and then left for about 30 seconds. When the beaker is again shaken with a rotatory motion the particles of rice move about freely if they are hand-pounded, but stick together, or to the bottom of the beaker, if milled.

ANNUAL REPORT OF THE CHEMICAL EXAMINER FOR THE YEAR 1930

THE Chemical Examiner's Department, which is a branch of the Education and Public Health Department, is under the direction of Dr. S. Rajagopal Naidu, M.B.E., M.B., B.S., F.I.C. The work comprises toxicological investigations, stain cases and miscellaneous medico-legal work, including the examination of firearms, bones and tissues, counterfeit coins, stamps, documents, etc. The total number of articles examined was 9392, as compared with 8913 in 1928.

HUMAN POISONING CASES.—Of the 458 cases investigated, poison was detected in 250, comprising 91 inorganic and 159 organic poisons. Oleander headed the list with 30 cases and datura and opium came next, with 29 and 25 cases respectively. Alcohol was found in 16 exhibits, aconitine in 18, madar juice in 13, nux vomica in 5, and opium in 6. Among the inorganic poisons, copper sulphate was identified in 27 cases, potassium or sodium nitrite in 13, mercury in 17, arsenic in 14, sulphuric acid in 9, and cyanide in 5.

Holarrhena antidysenterica, Wall.—This plant, commonly known as "kurchi" or "conessi," is a small deciduous tree with white flowers growing throughout the forests of India. Recent clinical experience has shown that it is a valuable antidiysenteric drug. It contains alkaloids which are known to be poisonous. Two children were given the bark of this plant as a purgative, and both died. The post-mortem examination revealed cardiac failure, the heart being distended with dark fluid blood. The specimen of the plant submitted yielded an alkaloidal extract, which killed frogs with paralysis and gave the following chemical reactions: (1) with sulphomolybdic acid a dark green colour; (2) with sulphovanadic acid a greenish-brown colour changing to purple at the edges; (3) with formaldehyde and sulphuric acid (Marquis' reagent) a reddish-brown colour turning blue at the edges. An alkaloidal extract giving these reactions was also obtained from the viscera of each of the children.

Nitrites.—Both potassium and sodium nitrite are extensively used by weavers in the dyeing of cloths in villages and can be bought without restriction. Cases of nitrite poisoning have therefore become increasingly frequent in recent years. The fatal dose for frogs is about 1 mg. of nitrite ion per 10 g. of body weight, and the fatal dose for dogs is similar, whether injected or administered by the mouth. The fatal dose for an average adult man, calculated by Meeh's formula (cf. *Extra Pharmacopoeia, Martindale*, 21st Ed., Vol. I, p. xxxii), would probably be 30 grains of sodium nitrite or 40 grains of potassium nitrite. A regular feature in animals killed by nitrite poisoning is the presence of methaemoglobin in the blood taken from the heart after death. When the poison is administered by the mouth most of it is rapidly absorbed and otherwise destroyed, and very little is left in the viscera. Hence in cases of human poisoning only small quantities of nitrite can be detected in the viscera unless very large doses have been swallowed.

ANIMAL POISONING CASES.—Of the 23 cases investigated, poison was detected in 13. Arsenic was found in 5 cases, oleander in 6, madar juice in 1, and a mixture of arsenic, mercury, copper, nux vomica seeds, aconitine, datura seeds and castor seeds in 1.

TATTOO MARKS.—Among the miscellaneous cases was one in which a man, accused of murder, was alleged to have cut the body of the victim into several pieces and to have buried them in a river bed. Some of these portions were recovered by the police, and on one of these pieces there were letters which appeared like tattoo marks corresponding to the name of the deceased. The problem submitted was to decide whether these letters were tattoo marks or merely writing in ink.

The characters were not affected by water, acetic acid, oxalic acid, ammonia, bleaching powder or potassium cyanide. A cross section of the skin showed dense fibrous tissue with superficial intracellular dark brown amorphous pigment. It was therefore concluded that the letters on the skin were tattoo marks and not surface writing.

ALTERATIONS ON A DOCUMENT.—A village officer was ordered to hold the elections for a panchayat board. It was suspected that he had not held these elections, but had reported falsely that he had done so on September 28th, 1938, and had subsequently tampered with the notices by altering the date and day of the week. Examination of the document by ordinary and ultra-violet light showed two places in ink of a deeper shade than on the remainder of the paper. It was also revealed that the figures and word of the date overlapped those of a date which had originally been November 5th, 1938.

RE-USED POSTAGE STAMPS.—Partly erased and re-used postage stamps are frequently submitted for identification of the erased postal impression. For this purpose Cellophane paper is very useful. The postal impression is printed on the transparent paper and can be superimposed on the suspected stamp. A complete coincidence of the circles and other marks that have not been erased enables a conclusion to be drawn.

Bibliography on Metals in Foods and Biological Materials

(Supplementing the series published in the ANALYST up to 1933, 58, 340, and bringing the Bibliography up to date.)

II. NICKEL

- ASH. Metals in Wineries. *Ind. Eng. Chem., Anal. Ed.*, 1935, 27, 1243; Abst., *B.C.A.*, 1936, 119B. (Protective coating forms on nickel apparatus.)
- BERTRAND and NAKAMURA. Investigation of the Physiological Importance of Nickel and Cobalt. *Bull. soc. sci. hyg. aliment.*, 1936, 24, 338; Abst., *C.A.*, 1937, 31, 4385.
- BERTRAND and NAKAMURA. Physiological Importance of Nickel and Cobalt. *Ann. Inst. Pasteur*, 1934, 53, 371; Abst., *B.C.A.*, 1934, 1395A.
- BEYTHIEN. Metal Utensils in Food Industries and in Kitchens. *Chem.-Ztg.*, 1936, 60, 107; Abst., ANALYST, 1936, 61, 263. (Nickel preferred to copper in German preserved food industries; small, non-toxic, amounts dissolve in foods.)
- BURRELL. Metals used in the Modern Dairy. *J. Milk Tech.*, 1938, 1, 25; Abst., *B.C.A.*, 1939, 204B. (With milk at high temperature, pure nickel unsatisfactory.)
- CHARLEY. Action of Cider on Metals. *Ann. Rept. Agr. Hort. Res. Sta.*, Long Ashton, Bristol, 1933, 152; Abst., *B.C.A.*, 1935, 249B. (No action on nickel.)
- CHOAG and NETTER. (Micro-) Photographic Records of Crystallised Insulin. *Presse méd.*, 1939, 47, 667; ref., *C.A.*, 1939, 33, 8360. (Zinc, nickel and cobalt compounds.)
- CLAYTON. Detection of Metals in Fibrous Materials, Dyes and Organic Pigments. *J. Soc. Dyers & Colorists*, 1937, 53, 380; Abst., *B.C.A.*, 1937, 1327B, and *C.A.*, 1938, 32, 360.
- CORMIA and STEWART. Nickel Dermatitis: A Report of Eleven Cases. *Canad., Med. Assoc. J.*, 1935, 32, 270; Abst., *C.A.*, 1935, 29, 3401.
- DIXON. Value of Nickel Salts in Treatment of Morton Mains Ailment. *New Zealand J. Sci. Tech.*, 1937, 19, 326; Abst., *B.C.A.*, 1938, 831B.
- FISHER and SCOTT. Ash Content of Nickel-insulin Crystals. *Trans. Roy. Soc. Canada*, 1938, 32, V, 55; Abst., *C.A.*, 1939, 33, 1442. (Nickel-insulin, from ash-free insulin, contained about 0.41 per cent. of nickel.)
- GENIN. Use of Metals in the Dairy Industry. II, Nickel and its Alloys. *Lait*, 1938, 18, 785; Abst., *B.C.A.*, 1939, 95B, and *C.A.*, 1939, 33, 246. (Review.)
- GODDEN. "Trace" Elements in Human and Animal Nutrition. *Chem. and Ind.*, 1939, 791; Abst., *B.C.A.*, 1940, 231AIII, and *C.A.*, 1939, 33, 8746. (Origin and physiological properties; nickel mentioned.)
- HELLER, KUHLA and MACHEK. Determination of Traces of Heavy Metals in Mineral Waters. *Mikrochem.*, 1937, 23, 78; Abst., ANALYST, 1938, 63, 374. (Polarographic; nickel mentioned.)
- HESS, OWENS and REINHARDT. Analysis of Organic Materials for Traces of Metallic Impurities. *Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 646; Abst., *C.A.*, 1940, 34, 961. (Spectroanalytical detection in biological material; nickel mentioned.)
- KONISHI and TSUGE. Mineral Matters of certain Leguminous Crops. I, Inorganic Constituents of Underground Parts of certain Leguminous Crops. *Mem. Coll. Agr., Kyoto Imp. Univ.*, No. 37 (*Chem. Ser.*, No. 20), 1936, 1; Abst., *C.A.*, 1936, 30, 4541. (Traces detected spectroscopically; nickel mentioned.)
- LA QUE. Nickel and its Alloys in Food Manufacture. *Food Ind.*, 1935, 7, 580; Abst., *B.C.A.*, 1937, 449B, and *C.A.*, 1936, 30, 525. (Non-corrosive in food-processing.)
- LEWIS. Spectrographic Determination of Nickel in Organic Material. ANALYST, 1935, 60, 13. (Arc method: Ratio quantitative system.)

- MOHR. "Emery" flavour in Milk and the Influence of Metals on this Defect. *Molherer-Zig.*, 1936, 50, 1454; Abst., C.A., 1937, 31, 1108. (Sometimes caused by nickel-lined containers.)
- MOHR and EICHSTAEDT. Action of Cream and Butter-fat on Metals, and the Influence of Metals on Quality of Butter and Clarified Butter. *Proc. World's Dairy Congress*, Milan, 1934, Sect. II, 95; Abst., B.C.A., 1934, 1080b, and C.A., 1934, 28, 5545. (Nickel attacked; butter quality impaired.)
- NAVES. Detection and Determination of Traces of Metals in Essential Oils, Solid Essences and Enflourage Products. *Parfums de France*, 1934, 12, 89, 116 and 139; Abst., C.A., 1934, 28, 5928. (Detection: acid extraction; determination: nitric-sulphuric combustion.)
- PAULAIS. Distribution of Nickel in Organs of Lamellibranch Molluscs. *Compt. rend.*, 1936, 203, 685; Abst., B.C.A., 1937, 7AIII; C.A., 1937, 31, 469. (Modification of Rollet's method; detects 1 to 10 γ in 10 ml. Most in branchiae and haematopaneas; least in muscle.)
- PIERCE, TORRES and MARSHALL. Qualitative Spectrographic Analysis, with Graphite Electrodes. *Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 41; Abst., B.C.A., 1940, 172AI. (Sensitivity for trace elements in biological ash in spectral regions 2500 to 3400Å given: Nickel 0.40 γ .)
- PRAT. Detection of Heavy Metals in Plants and the Chromosopodogram Method. *Mikrochem., Molisch Festschr.*, 1936, 342; Abst., B.C.A., 1937, 243AIII. (Nickel detected microscopically with dimethylglyoxime.)
- RAMAGE. Analysis of Tissue for Metallic Content. *Nature*, 1936, 138, 762; Abst., B.C.A., 1936, 1532A; C.A., 1937, 31, 1061. (Extraction with N hydrochloric acid before spectrographic determination; nickel mentioned.)
- RAMAGE. Biological Distribution of Metals. *Nature*, 1936, 137, 67; Abst., B.C.A., 1936, 258A, and C.A., 1936, 30, 2605. (Spectroscopic determination in foods. Nickel:—St. Ignatius beans, 0.014 per cent.; many spices and herbal drugs, traces; indiarubber tree seeds, from 0.001 per cent. [endosperm] to nil [outer shell]; tea.)
- RANEDO. Source of Error in the Determination of Nickel in Hydrogenated Oils. *Anal. Fts. Quim.*, 1935, 33, 813; Abst., C.A., 1936, 30, 3264, and B.C.A., 1936, 846B. (Ferrous iron from plant gives red colour with dimethylglyoxime.)
- RIGG. Bush Sickness in Sheep: Mineral Content of Pasture Investigation at the Cawthron Inst., 1935–6. *Tenth Ann. Rept. Dept. Sci. Ind. Res., New Zealand*, 1936; Abst., C.A., 1937, 31, 5024. (Nickel administration [8 mg./week] improved health but did not always cure.)
- ROSENBERGER. Action of Alcoholic Solutions of Essential Oils on Various Metals. *Seifensieder-Zig.*, 1937, 64, 967; Abst., B.C.A., 1938, 319B. (Nickel not attacked by peppermint, lavender and lemon oils.)
- ROST. Concentration of the Chemical Elements by Organisms. *Pyroda*, 1936, 29, 223; Abst., C.A., 1939, 33, 653. (Concentrations in plant and animal tissues summarised; nickel mentioned.)
- SAHYUN, NIXON and GOODELL. Influence of certain Metals on the Stability of Insulin. *J. Pharm. Exp. Ther.*, 1939, 65, 143; Abst., B.C.A., 1939, 506AIII. (Nickel [1 mg./1000 units] preserves insulin activity for 7 weeks.)
- SHELDON. Mineral Basis of Life. *Brit. Med. J.*, 1934, I, 47; Abst., C.A., 1934, 28, 2035. (Review of physiological effects of metals; nickel mentioned.)
- STEWART. Inherent Sensitivity of the Skin to Nickel and Cobalt. *Arch. Int. Med.*, 1933, 51, 427; Abst., B.C.A., 1933, 532A.
- STRUSZYŃSKI. Detection of Traces of Nickel in Hydrogenated Fats. *Przemysł Chem.*, 1935, 19, 48; Abst., B.C.A., 1935, 508B. (Glyoxime spot method.)
- SULLIVAN. Inorganic Constituents of Wheat and Flour. *Cereal Chem.*, 1933, 10, 503; Abst., C.A., 1934, 28, 1412. (Nickel in wheat 35 p.p.m., in patent flour and in bread nil.)
- TORRICELLI. Identification and Determination of Traces of Nickel in Hydrogenated Fats. *Mitt. Lebensm. Hyg.*, 1937, 28, 36; Abst., B.C.A., 1937, 1078B. (Colorimetric [dimethylglyoxime] determination after removal of iron from acid extract.)
- UNDERWOOD. Significance of the "Trace" Elements in Nutrition. *Nutrition Abs. and Rev.*, 1940, 9, 515. (Nickel mentioned.)
- WEBB. II. Spectrographic Analyses of Marine Invertebrates, with special Reference to Chemical Composition of their Environment. *Sci. Proc. Roy. Dublin Soc.*, 1937, 21, 505; Abst., C.A., 1937, 31, 6268. (Nickel mentioned.)
- WHITFIELD, DAVIES and DOWNS. Effect of Milk upon Metals and Metals on Milk. *Milk Dealer*, 1934–5, 24, 34, 40, 42; Abst., C.A., 1935, 29, 5528. (Nickel attacked; milk "off" flavour only when corrosion great.)

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Hydrogen Sulphide in Corned Beef.

A. Elias. (*Indust. y. Quim.*, 1940, 3, 31-32.)—The presence of small amounts of hydrogen sulphide in Argentine corned beef has been reported. To discover its origin, 20 g. of the samples were treated with 50 ml. of 10 per cent. sulphuric acid, and the hydrogen sulphide evolved was estimated colorimetrically by comparing the stains produced on lead acetate paper in the course of 24 hours with standards prepared from sodium sulphide of known purity. It was found that a faint reaction was obtained with 0.00187 mg. of hydrogen sulphide, and this was taken as the lowest limit. The most constant results were obtained by using only 1 g. of the sample of beef. Freshly killed meat yielded traces of hydrogen sulphide (about 0.05 mg. per 100 g.), but this disappeared after the sample had been kept in a refrigerator for 48 hours. On the other hand, a sample of corned beef which had been sterilised for 2½ hours at 113° C. yielded 3.74 mg. per 100 g. It is recommended that an effort should be made to reduce the decomposition of the proteins to a minimum, so that the amount of hydrogen sulphide should not exceed the 0.05 mg. per 100 mg. present in the fresh meat.

Detection of the Bleaching of Flour with Chlorine.

D. B. Scott. (*J. Assoc. Off. Agr. Chem.*, 1940, 23, 675-678.)—The following method for the determination of chlorine in the fat of flour has been proposed (Scott, *J. Assoc. Off. Agr. Chem.*, 1940, 23, 497). The flour (500 g.) is shaken at five-minute intervals for 30 minutes with 700 ml. of petroleum spirit, the mixture is filtered by means of a Buchner funnel, and the filtrate is passed through the flour a second time to ensure freedom from suspended flour, and is then concentrated to 10 ml. and treated by one of the following methods. By the "quick ash method" the extract (10 ml.) is added to 15 g. of fusion mixture (138 g. of anhydrous potassium carbonate, 106 g. of anhydrous sodium carbonate and 75 g. of potassium nitrate) which is then dried and weighed to determine the amount of fat added. A further 5 g. of fusion mixture is spread over the surface, and the mixture is ignited to a white ash at 525° to 600° C. The residue is treated with 25 ml. of hot water and rinsed into a 200-ml. beaker with hot water, acidified to litmus paper with nitric acid, treated with a further 25 ml. of nitric acid and 5 ml. of 0.3 N silver nitrate solution and boiled for 5 minutes. The solution is filtered through a chlorine-free paper, which is washed with 1 per cent. nitric acid, and the A.O.A.C. method is then followed, beginning at the words "place the paper in a Kjeldahl flask. . . ." By the "acid digestion

method" the extract is evaporated to dryness, the fat is weighed and treated with 5 ml. of 0.3 N silver nitrate solution and 25 ml. of nitric acid, and the volume of liquid is reduced to about 10 ml. While the liquid is briskly boiling, nitric acid is added, a few ml. at a time, until the fat is completely destroyed, after which an equal volume of water is added and the liquid is boiled for 5 minutes and filtered. The A.O.A.C. method is then followed from the point previously indicated. The fat of the flour should be subjected to a preliminary Beilstein test for chlorine, and if this indicates a high content the amount of sample should be reduced to 200 g., and 300 ml. of petroleum spirit should be used for extraction. The application of the method to the determination of known amounts of sodium chloride added to 4 g. of olive oil showed that no chlorine is lost in the process. The chlorine found in the fat of unbleached flour ranged from 0.03 mg. to 0.19 mg. per g. of fat. Flours bleached with chlorine and nitrosyl chloride gave 10.3 to 18.9 mg. per g. From the results it may be concluded that chlorine in excess of 0.25 mg. per g. of fat indicates that the flour has been treated with bleaching preparations. The average chlorine-content of 28 authenticated samples of unbleached flour was 0.11 mg. per g. of fat.

A. O. J.

Copper in Tomatoes.

W. J. Shannon and D. T. Englis. (*J. Assoc. Off. Agr. Chem.*, 1940, 23, 678-680.)—Samples of several varieties of tomatoes, obtained from different growers in central and northern Illinois and subject to the effect of different types of soil and other factors, were examined for their copper-content. The tomatoes were washed twice with distilled water, rinsed once with copper-free water, and then pulped in a food chopper, only the stems being excluded. The material was prepared for analysis by oxidation with nitric and perchloric acids (Gieseking, Snider and Getz, *Ind. Eng. Chem., Anal. Ed.*, 1935, 7, 185). The copper was estimated with the sodium diethyldithiocarbamate reagent, a single cell photoelectric colorimeter and a blue filter being used, as recommended by Drabkin (*J. Assoc. Off. Agr. Chem.*, 1939, 22, 320). The calibration curve in the range of concentration studied was constructed by means of solutions of pure copper sulphate. The results showed that the average copper-content of tomatoes from these regions ranged from 15 to 25 parts per million.

A. O. J.

Macaúba Oil (Mocaya).

M. Silva. (*Indust. y. Quim.*, 1940, 3, 39-41.)—According to some authors, macaúba oil is derived from the palm *Acrocomia sclerocarpa*, but Bolton states (*Oils and Fatty Foods*, p. 176) that it is obtained from *Acrocomia totai*. A sample extracted

from nuts from Minas-Geraes gave the following values: sp.gr. at 15° C., 0.9212; n_D^{25} , 1.4631; m.p., 23.6° C.; solid. p., 18.3° C.; acid value, 3.40; saponification value, 239.70; iodine value (Hanus), 22.6; glycerol (calc.), 12.76.

Component Glycerides of a Typical Cow Milk Fat. T. P. Hilditch and S. Paul.

(*J. Soc. Chem. Ind.*, 1940, 59, 138-144.)—The method of separation of fats by preliminary crystallisation from acetone into one or more fractions, the component glycerides of which can be determined within fairly near limits, has now been extended to a typical butter-fat produced by cows at summer pasture. At least 4 main groups of acyl compounds have to be considered: palmitic, stearic, oleic with octadecadienoic and traces of unsaturated C_{20} and C_{22} acids, and the separate group of characteristic milk-fat lower saturated acids from butyric to myristic, with traces of decenoic, do- and tetra- decenoic acids, so that final deductions are less certain than with simpler fats and necessitate the grouping together of all the lower acids below C_{16} . The original butter-fat was separated into three fractions, and their component acids were determined by ester fractionation; the fully saturated glycerides were determined and isolated, and, when possible, fractionated by crystallisation and the component acids determined; the completely hydrogenated fraction was separated by crystallisation from acetone or ether followed by component acid analysis of each hydrogenated fraction obtained. The mixed glycerides (per cent. mol.) were probably in the neighbourhood of "oleo"-mono- C_{4-14} palmitins 31-22, "oleo"-palmitostearins 8-17, palmitodi-"oleins" 17-4, "oleo"-mono C_{4-14} stearins 12-6, smaller proportions of "oleo"-di- C_{4-14} 4-9, mono C_{4-14} di-"oleins" nil-10, stearodi-"oleins" 8-1, and "oleo"-dipalmitins, 1-5; tri-"oleins," nil-7 (very probably mainly octadecadienodiolein), and fully saturated glycerides (mono- C_{4-14} palmitostearins 9, di- C_{4-14} palmitins 6-7, and small amounts of di- C_{4-14} -stearins, mono- C_{4-14} -palmitins, dipalmito stearins and palmitodistearins) amounting in all to 19 per cent. mol. Glycerides containing one radical each of oleic, palmitic, and one of the lower acids are most abundant (22-30 per cent. of the whole fat), whilst nearly 40 per cent. of the fat is made up of the four groups "oleo"-mono- C_{4-14} -palmitins, "oleo"-palmitostearins, palmitodi-"oleins" and "oleo"-mono- C_{4-14} stearins, and 19 per cent. of fully saturated glycerides are present. Palmitic acid is combined with other acids in about 70 per cent. of the butter glyceride molecules, and there is a similar distribution in the depôt fat of the cow. It is suggested that butter-fat may be the result of transformation of pre-formed oleoglycerides in the mammary gland, and the relative distribution of the lower fatty acids and of stearic acid are discussed in this relation. D. G. H.

Detection of Adulteration in Ghee by a Study of its Fluorescence. G. Narasimhamurthy and V. V. Suryanaraynamurthy. (*Current Science*, 1940, 9, 334-336.)—Jha (*J. Ind. Chem. Soc., Ind. & News Ed.*, 1939, 1, 159) has described a qualitative test for the adulteration of ghee, which depends on its fluorescence in ultra-violet light, and Muthanna and Mukerji (*Current Science*, 1940, 9, 120) have evolved a similar quantitative method, in which a Pulfrich photometer is used to measure the intensity of the fluorescence and to obtain standard curves for the common adulterants of ghee. Earlier work has suggested that such methods might be influenced by the age of the sample and the presence of artificial colours. Tests were therefore made in which the intensity of the fluorescence produced by the ultra-violet light transmitted by filters that allow the passage of radiations of wave-lengths 520 to 540 and 450 to 490 $m\mu$ (which are regarded as characteristic of ghee and its adulterants) was measured in a Pulfrich photometer. A solution containing 880 γ of quinine per litre was used in a similar way as standard, and the solution tested contained 1 vol. of the melted fat and 3 vol. of chloroform. The results show that a combination of the blue fluorescence of crude groundnut oil and the yellow fluorescence of a colouring matter which is commonly used in the dairy industry for adding to butter, can simulate the greenish fluorescence associated with genuine ghee. The ratio of the intensities of the blue and green components of the fluorescence indicated no relationship with factors such as the age and purity of the sample. J. G.

Neutralisation Value of Ghee (Butter-fat). H. Hawley. (*Current Science*, 1940, 9, 337-339.)—The Reichert value is unsatisfactory as an indication of adulteration of ghee, because the herds of animals (usually buffaloes) from which it is obtained are subjected to very variable climatic and feeding conditions, and the composition of the butter-fat varies accordingly, resulting in Reichert values ranging from less than 20 to over 40. Tests which depend on the butyric acid content are also unreliable, because they do not make it possible to decide whether a low Reichert value is due to abnormality or to adulteration with a fat which contains no butyric acid. Other tests (e.g. the saponification value, the n_D or the sterol acetate test) also have their limitations. The neutralisation value (N.V.) of the fatty acids is of greater use if determined by the author's method, which minimises losses of volatile acids (e.g. caproic and caprylic acids); it does not include the butyric acid, which is soluble in water. About 10 g. of the clear fat are saponified with 150 ml. of water and 30 ml. of a solution of 84 g. of sodium hydroxide in 210 ml. of water, to which is added sufficient glycerol to make 1 litre. This mixture is heated on a water-bath in a 500-ml. flask fitted with an 8-inch air-condenser, the top of which is covered with an inverted test-tube. When hot, the contents of the flask are acidified with

10 ml. of approximately 17 per cent. (by vol.) sulphuric acid, the strength of which has previously been adjusted (as the result of a blank titration with methyl red as indicator), so that 9.0 to 9.5 ml. serve to neutralise the alkali present. After 3 hours (not more) on the water-bath, with frequent shaking, the fatty acids are clarified, and 50 ml. (which includes all or most of the fatty acids) are poured off and washed with three 25-ml. portions of hot water in a separating funnel. The acids are filtered (in a steam oven), and about 5 g. of the filtrate are pipetted into a tared flask and weighed (to within 2 mg.). They are then dissolved in 50 ml. of 90 per cent. alcohol (which has previously been neutralised to thymol blue), and the solution is titrated with a freshly made 0.5 N solution of sodium hydroxide, with 1 ml. of 0.04 per cent. thymol blue solution as indicator, until the yellow colour has changed to a slate-green shade. The *N.V.* is then the number of mg. of potassium hydroxide required to neutralise 1 g. of fatty acid; duplicates should agree to within 0.2. Results tabulated for 78 samples showed that, although there is some tendency for high Reichert values to be associated with high neutralisation values, they do not run parallel with one another in the way that the former does with the saponification value. It is suggested that a sample of bulked ghee which has a *N.V.* of less than 209 is almost certainly adulterated, and that if the *N.V.* is less than 210 the sample must be regarded with suspicion. Of 7 samples which gave a positive sterol acetate test, 5 could have been condemned on the *N.V.* alone, whilst one had a *N.V.* of 209 to 210, and there was reason to believe that adulteration of the other was only slight. Of 10 remaining samples, none of which contained vegetable fat, 2 were classed as adulterated, 1 as doubtful, and the others as genuine. Incidentally, a substitute for the sterol acetate test is an advantage in view of the present difficulty of obtaining digitonin. J. G.

Lower Saturated Fatty Acids in Herring Oil. H. Nobori. (*J. Soc. Chem. Ind. Japan*, 1940, 43, 110b.)—About a ton of the mixed fatty acids of herring oil (neutralisation value 197.3, saponification value 199.0, iodine value 128.9) was distilled in a commercial plant, and 28.5 per cent. of the lower-boiling fraction (205–217° C. at 12–10 mm.), with neutralisation value 221.1, saponification value 220.9 and iodine value 76.9, were obtained. This fraction contained lauric acid (2.2 per cent.) and capric and caprylic acids (0.9 per cent. of the original mixed fatty acids). Phytosteric acid ($C_{14}H_{26}O_2$) is regarded as a constituent of the unsaturated acids, and a hydrocarbon, pristane, as a main constituent of the unsaponifiable matter of the fraction of lower b.p. D. G. H.

Selective Action of Fatty Acid on the Ethanolysis of Sardine Oil. M. Takano, M. Takao and M. Danjo. (*J. Soc. Chem.*

Ind. Japan, 1940, 43, 132–133b.)—The refined oil was mixed with an equal volume of 0.5 N sodium hydroxide solution in 95 per cent. alcohol, the mixture was shaken vigorously at 20° C., and the resulting products were taken from the reaction mixture during the course of ethanolysis. The reaction ceased on addition of dilute hydrochloric acid, and some free fatty acids were liberated from the soap produced. Samples were washed with hot water and each was assumed to consist of tri-, di- or mono-glyceride, free fatty acids and ethyl ester. The products were then distilled under 2–3 mm. pressure to separate ethyl esters, and the samples showed increasing free fatty acidity; hence the alkali for saponification can be only a small portion of that used, and most of it probably acts as catalyst for the ethanolysis. The distillate and residue were also analysed. The high acetyl values of the residues show that di- or mono-glyceride may be produced in ethanolysis. The saponification value of the distillate is generally higher than that of the residue and the iodine value lower, so that the fatty acids of lower molecular weight and those of less saturation may react more rapidly. The distillate and residue were treated by the lithium salt and acetone method to separate unsaturated fatty acids, and the residue was further separated into solid and liquid fatty acids by the lead salt procedure. Results of analysis are given. Solid fatty acids react to the greatest extent, liquid (less unsaturated) acids come next, and highly unsaturated react least. Further, the saturated acids of lower molecular weight tend to react more rapidly than the other solid acids; of the highly unsaturated acids, and even of the liquid acids, those with high neutralisation values react more rapidly than the others. D. G. H.

Determination of Mercury in Mercurochrome. G. J. W. Ferrey. (*Pharm. J.*, 1940, 144, 366.)—In the B.P. method the mercury in mercurochrome is determined in acid solution, and a large proportion of alcohol is required to keep dissolved the dibromohydroxymercurifluorescein that is formed; the results may be low and erratic. The following method, which can be carried out in 30 minutes, gives consistent and accurate results:—A weighed quantity (0.5 to 1 g.) of the sample is dissolved in 20 ml. of water, and the solution is treated with 5 g. of potassium hydroxide pellets and 2 g. of zinc filings and boiled under reflux for at least 15 minutes. The condenser is then washed down with 50 ml. of water, and the amalgam is separated by decantation, washed well with water, and dissolved in a mixture of 20 ml. of nitric acid and 20 ml. of water. The solution is gently boiled until nitrous fumes cease to be evolved, treated with a slight excess of permanganate, decolorised with a drop of hydrogen peroxide solution, and titrated with *N*/10 ammonium thiocyanate solution.

The Bambarra Groundnut or Njugo Bean. J. M. Holm and B. W. Marloth. (*Dept. of Agric. and Forestry, South Africa, Pamphlet No. 215, 1940, 1-10*).—The Njugo bean (*Voandzeia subterranea*) is widely cultivated throughout tropical Africa, and Bambarra (a district near Timbuctoo) has no pre-eminent claim to the plant. It is also extensively grown in the Transvaal, Natal and Zululand. It resembles the ground-nut (*Arachis hypogaea* L.) only in so far as it matures its seed in a pod underground. Several varieties, distinguished mainly by the colour of the shelled seed, are recognised by the natives. An analysis of the dehusked beans by D. C. Crawford gave the following results: moisture, 10.3; protein ($N \times 6.25$), 15.0; fat, 7.4; fibre, 5.1; carbohydrates, 59.1; ash, 3.1; phosphoric oxide, 0.6; potash, 1.1. The following table gives the percentage composition of green and ripe nuts, as determined at the Station Agronomique, Mauritius.

| | Water | Protein ($N \times 6.25$) | Fat | Fibre | Carbohydrate | Ash |
|--------------------------|-------|--------------------------------|------|-------|--------------|------|
| Nuts (green) | 58.5 | 7.3 | 3.2 | 3.0 | 26.4 | 1.16 |
| Shells (green) | 80.3 | 1.6 | 0.1 | 5.5 | 11.7 | 0.8 |
| Entire fruits (green) .. | 63.6 | 6.02 | 2.43 | 3.58 | 23.13 | 1.46 |
| Nuts (ripe) | 12.5 | 14.7 | 6.1 | 5.7 | 57.1 | 3.9 |
| Shells (ripe) | 11.1 | 7.3 | 0.8 | 29.1 | 47.8 | 3.9 |
| Entire fruits (ripe) .. | 12.27 | 13.36 | 5.17 | 9.86 | 55.4 | 3.94 |

Composition of Certain Nutshells. M. Phillips and M. J. Goss. (*J. Assoc. Off. Agr. Chem.*, 1940, **23**, 662-665).—The percentage compositions of the following nutshells were determined:—Almond (*Amygdalus communis*), Brazil nut (*Bertholletia excelsa*), candle nut (*Aleurites moluccana*), coconut (*Cocos nucifera*), English walnut (*Juglans regia*), filbert (*Corylus avellana*), and pecan (*Hicoria pecan*). The shells were ground to pass through a 60-mesh sieve and dried at 105° C. Ash was determined by ignition at 600° C., nitrogen by the Kjeldahl-Gunning method, methoxyl by the method previously described (Phillips, *J. Assoc. Off. Agr. Chem.*, 1932, **15**, 123; Abst., ANALYST, 1932, **57**, 402), and extractives by the method of Phillips *et al.* (*J. Agr. Res.*, 1939, **59**, 319). Uronic acid anhydrides were determined in the unextracted material by the method of Dickson, Ollerson and Link (*J. Amer. Chem. Soc.*, 1930, **52**, 775) as modified by Phillips, Goss and Browne (*J. Assoc. Off. Agr. Chem.*, 1933, **16**, 289; Abst., ANALYST, 1933, **58**, 495), furfuraldehyde by the method of the A.O.A.C. and crude cellulose by the method of Kürschner and Hanak (*Z. Unters. Lebensm.*, 1930, **59**, 484). Pentosans and lignin were determined by methods previously described (*cf. refs. pp. 324, 326, J. Assoc. Off. Agr. Chem.*, 1940, **23**). The figures found for ash and nitrogen were low except for candle nut (ash, 3.64 per cent.). The percentages of methoxyl were somewhat higher than those found in lignified plant materials, and ranged from 5.29 (almond) to 7.14 (walnut). The percentages of alcohol-benzene extractives were rather low, but those of other extractives as well as of the uronic acids were of the order

usually found for lignified substances. The percentages of pentosans were generally higher in materials of low lignin-content and *vice versa*, although this relation was neither regular nor proportional. The percentages of lignin in candle nut (37.29) and pecan (35.51) were unusually high, higher even than those for such highly lignified material as woods (*cf. Fleck, Van Beckum and Ritter, J. Amer. Chem. Soc.*, 1937, **59**, 2279; Abst., ANALYST, 1938, **63**, 66). Where the percentage of lignin was high the percentage of methoxyl in lignin was somewhat low (*e.g.* candle nut 12.03; pecan, 11.03). Almond shells with a low percentage of lignin (17.13) gave 17.54 per cent. of methoxyl in "pure" lignin. The figures for cellulose and crude cellulose showed little variation in the kinds of shells examined.

A. O. J.

Phenol and Eucalyptus Ointments. H. Brindle. (*Pharm. J.*, 1940, **145**, 20).—Experiments on the preparation and storage of

these ointments show that the loss of phenol in preparing about 250 g. of phenol ointment by ordinary methods is approximately 5 per cent. If the ointment is stored in collapsible tubes the loss is negligible, even after a year or more; in well-closed containers the loss in 9 months may reach about 8 per cent., but it can be kept down to much less. The loss in preparation is about 1 to 2 per cent. at 60° C. and 5 per cent. at 80° C. In full, well-closed containers the loss of oil on storage for 18 months and for 3 years was only about 2 to 3 per cent. It is concluded that the most satisfactory way of dealing with ointments containing volatile ingredients is to purchase them in collapsible tubes. Such ointments should not be stored in the usual shop jar with a loosely fitting cover. If kept in airtight containers, particularly if well filled, there should be no danger of trouble with the authorities if the samples are not stored for more than 1 or 2 years. The ointments may be prepared freshly as required. It is possible that the efficiency of collapsible tubes is not altogether due to their being airtight, as stoppered bottles show a loss, but largely due to complete filling preventing condensation on the sides of the container.

E. M. P.

Biochemical

Determination of Carotene in Presence of Lycopene. G. S. Fraps, A. R. Kemmerer and S. M. Greenberg. (*J. Assoc. Off. Agr. Chem.*, 1940, **23**, 422-425).—Impurities occurring in crude carotene solutions can be removed to some extent by means of magnesium carbonate specially prepared so as to adsorb

crude xanthophyll but not carotene, and called the xanthophyll reagent (*cf.* Fraps and Kemmerer, *J. Assoc. Off. Agr. Chem.*, 1939, **22**, 190; *Abst.*, *ANALYST*, 1939, **64**, 369). Some foods (*e.g.* tomato, water melon, red pepper) contain lycopene or other red pigments which are not completely adsorbed by the xanthophyll reagent. Experiments were made to devise a method of preparation of a reagent that would adsorb lycopene but not carotene. A test solution of crude lycopene was prepared by refluxing 100 g. of canned tomato pulp for 30 minutes with 250 ml. of saturated alcoholic potassium hydroxide solution. The carotene fraction, containing lycopene, was extracted by means of petroleum spirit as in the Hughes Peterson modification of the Guilbert method (*J. Assoc. Off. Agr. Chem.*, 1939, **22**, 79), and the extract was washed with 90 per cent. methanol to remove xanthophyll, then with water to remove residual methanol, and finally dried over sodium sulphate. The solution was diluted to 200 to 400 ml. with petroleum spirit and shaken with 50 g. of the xanthophyll reagent. The filtrate was treated gradually with a preliminary preparation of magnesium carbonate that would adsorb lycopene without adsorbing carotene. The adsorbent was filtered off and washed with petroleum spirit, and the purified lycopene was eluted from the magnesium carbonate with petroleum spirit containing 2 per cent. of ethanol. The eluate was washed free from alcohol, dried over sodium sulphate, and made up to a colour strength equivalent to 0.5 to 0.8 p.p.m. of carotene. Lycopene solutions thus prepared are unstable, even when kept in the dark in cold storage, and should be freshly prepared every week. To test the reagent, the colours of test solutions of lycopene and carotene were read in a KWSZ photometer before and after treatment of 50 ml. of each with 3 to 5 g. of the reagent. To be satisfactory the reagent must adsorb all the lycopene from a solution containing 0.5 to 0.8 p.p.m. of lycopene, and no carotene from a solution containing the same amount of carotene. Xanthophyll will also be adsorbed by the lycopene reagent. After a number of trials a lycopene reagent was prepared as follows:—Magnesium carbonate (100 g.) was heated in an electric furnace at 200° C. for 1 hour and then tested for adsorption of carotene. If the reagent did not adsorb carotene it was tested with the lycopene solution, and, if 3 to 5 g. of the reagent extracted all the red pigment from 50 ml., it was ready for use. If the reagent adsorbed carotene, water was added in 3-ml. portions until there was no further adsorption. It was then tested with the lycopene solution. Some modification of the procedure (*e.g.* longer heating) may be necessary with different specimens of magnesium carbonate. The lycopene reagent removed 76.7 per cent. of impurity from crude carotene in solutions derived from water melons, compared with 8.3 per cent. by the xanthophyll reagent. From carotene derived from dried apricots the lycopene reagent removed 31 per cent. of impurity and the

xanthophyll reagent 8.6 per cent. The crude carotene from tomato, red pepper and alfalfa also yielded more impurity to the lycopene reagent than to the xanthophyll reagent.

A. O. J.

Adsorption Method for the Determination of Pure Carotene. G. S. Fraps, A. R. Kemmerer and S. M. Greenberg. (*J. Assoc. Off. Agr. Chem.*, 1940, **23**, 659-662.)—In the tentative method of the A.O.A.C. for the determination of carotene (*J. Assoc. Off. Agr. Chem.*, 1939, **22**, 79) the material is boiled with alcoholic potassium hydroxide solution, petroleum spirit is added, the resulting mixture is diluted with water and the petroleum spirit layer is removed and washed with water and then with dilute methyl alcohol. The method might be shortened by treating the petroleum spirit extract with xanthophyll adsorbent (*cf.* preceding abstract) immediately after it has been washed with water, the washing with methyl alcohol being omitted. An investigation of this procedure showed that more washings with water were required to remove the alkali than were necessary when methyl alcohol was used, but that the alkali could be effectively removed by a final washing with dilute hydrochloric acid. The method finally used was as follows:—The sample (1 to 6 g.) was extracted with 20 to 120 ml. of saturated alcoholic potassium hydroxide solution as in the A.O.A.C. method (*loc. cit.*). The extract was diluted, and the carotinoid pigments were extracted with petroleum spirit, which was washed three times with water and once with a 0.5 per cent. solution of hydrochloric acid. The solution was dried over anhydrous sodium sulphate and diluted to 200 ml., and about 100 ml. were shaken for one minute with 8 g. of the xanthophyll adsorbent. The clear solution was examined in a photoelectric colorimeter, and the solution was again treated with 4 g. of the adsorbent, the treatment being repeated until no more colour was removed. If the material contained lycopene, the lycopene adsorbent was used (*cf.* the preceding abstract). The method was applied to 19 samples, and the shorter method gave the same results as the longer modified A.O.A.C. method. A. O. J.

Colorimetric Assay of Weakly Phenolic Ketones, "Oestrone," in Extracts of Human Urine. N. B. Talbot, J. K. Wolfe, E. A. MacLachlan, F. Karush and A. M. Butler. (*J. Biol. Chem.*, 1940, **134**, 319-330.)—A 24-hour sample of urine, containing 7 ml. of hydrochloric acid as preservative, is hydrolysed, within 12 hours of collection, by boiling under reflux for exactly 10 minutes with 15 per cent. of hydrochloric acid by vol. The liquid is then cooled and extracted 4 times with one-fifth of its volume of ether. The combined ethereal extracts are washed with 20 ml. of 20 per cent. sodium carbonate solution, evaporated to 100 ml., and transferred quantitatively to a separating funnel; the volume is adjusted to about 190 ml. The ethereal solution is washed alternately with

three 25-ml. portions of 0.1 *N* sodium hydroxide solution and two 25-ml. portions of 0.1 *N* sodium hydroxide—sodium hydrosulphite reagent (prepared immediately before use by adding 50 ml. of 0.1 *N* sodium hydroxide solution to 5 g. of sodium hydrosulphite) to remove strongly acidic and phenolic substances. In extracting with the hydrosulphite solution the funnel should be shaken thoroughly for at least 3 minutes. The ether is next washed twice with 25-ml. portions of 0.5 *N* hydrochloric acid and three times with 25-ml. portions of water; all the washings are discarded. The ethereal solution is then evaporated almost to dryness, and the residue is transferred to a 250-ml. separating funnel by means of four 25-ml. portions of toluene. The weakly phenolic substances are extracted from this solution with four 25-ml. portions of *N* sodium hydroxide solution, and the combined extracts are made acid to Congo red with hydrochloric acid and then adjusted to a *pH* that is acid to litmus and alkaline to Congo red by adding 20 per cent. sodium carbonate solution. The solution is extracted with five 40-ml. portions of ether, and the combined ethereal extracts are washed successively with 25 ml. of 0.1 *N* sodium hydroxide solution, 2.5 ml. of 0.1 *N* sodium hydroxide—sodium hydrosulphite reagent (shaking for at least 3 minutes), 25 ml. of 0.1 *N* sodium hydroxide solution, two 25-ml. portions of 0.5 *N* hydrochloric acid and three 25-ml. portions of water. The ethereal solution is evaporated to dryness, and the residue is transferred with four 1-ml. portions of absolute alcohol to a small Pyrex vessel graduated at 4 ml. and fitted with an all-glass reflux condenser. After addition of 0.5 ml. of glacial acetic acid and 0.5 g. of Girard's reagent T (betaine hydrazide hydrochloride, stored in a vacuum desiccator over conc. sulphuric acid) to the alcoholic solution, it is heated under reflux for half-an-hour and, after cooling, transferred with the aid of 40 ml. of ice-cold water to a separating funnel. Three ml. of 10 per cent. sodium hydroxide solution are added, and the solution is extracted with four 40-ml. portions of ether; the ethereal extracts are discarded. The aqueous solution is run into a flask containing 1 ml. of conc. sulphuric acid diluted with 80 ml. of water, and the liquid is transferred to a separating funnel. Forty ml. of ether are added, and the mixture is allowed to stand at room temperature for 75 minutes and then shaken. The ethereal extract is removed, and the aqueous solution is re-extracted with four further 40-ml. portions of ether. The combined ethereal extracts are washed with 25 ml. of 0.1 *N* sodium hydroxide solution, followed by three 25-ml. portions of water, and evaporated to dryness, and the residue, containing the weakly phenolic ketones, is dissolved in 1 to 4 ml. of absolute alcohol. An aliquot portion (0.3 ml.) of the solution is transferred to a small dry test-tube and mixed with 0.75 ml. of 2 per cent. sodium carbonate solution. Wetting of the upper walls of the test-tube must be avoided during this operation. A solution

(0.1 ml.) of diazotised dianisidine* is added as rapidly as possible, with stirring, followed one minute later by 0.8 ml. of toluene. The tube is shaken vigorously and then centrifuged together with a blank prepared in the same way, but containing no hormone. Aliquot portions (0.6 ml.) of the clear coloured toluene solutions are transferred from the tubes to the 1-ml. cells of a photoelectric colorimeter, and the galvanometer readings are taken with filter 420. The results (in γ of weakly phenolic ketones, "oestrone") are calculated either from a standard curve prepared with solutions of crystalline oestrone of known dilution, or from the expression $(2 - \log G)/K$, where *K* is the "proportionality constant" determined from solutions containing known amounts of crystalline oestrone and *G* is the galvanometer reading of the unknown. Crystalline oestrone can be estimated with an error of approximately 10 per cent. An average recovery of 72 per cent. of crystalline oestrone in pure solutions was obtained in the purification of extracts up to treatment with Girard's reagent T, and an average recovery of 95 per cent. of crystalline oestrone in the ketonic fraction after treatment with that reagent. When crystalline oestrone was added to the crude ethereal extracts of hydrolysed urine, recoveries averaging 65 per cent. were obtained, comparable with an average recovery of 67 per cent. with pure solutions for the complete purification procedure.

F. A. R.

Thiochrome Method for the Estimation of Aneurin, with Survey of the Aneurin Content of Wheats. R. G. Booth. (*J. Soc. Chem. Ind.*, 1940, 59, 181-184r.)—Pyke's method (this vol., 180) is considered to be the most suitable modification of Jansen's reaction (*ANALYST*, 1937, 62, 60) for the estimation of aneurin in wheat, but it is open to

* To 10 g. of crude dianisidine (m.p. 134.5 to 136.5° C.) are added 140 ml. of water and 6 ml. of conc. hydrochloric acid, and the mixture is boiled until the solid dissolves completely. The boiling is continued for a further 5 minutes after addition of 0.5 g. of stannous chloride, and the solution is finally decolorised by adding 1 g. of Norit A. The filtrate is treated with 60 ml. of conc. hydrochloric acid and cooled, and the purified dianisidine dihydrochloride, m.p. 283° C. (decomposition), is filtered off, washed with ethyl alcohol, and dried in a vacuum desiccator over sodium hydroxide until free from hydrochloric acid. Three ml. of water, followed by 0.3 ml. of conc. hydrochloric acid, are added to 25 mg. of dianisidine dihydrochloride, and the solution is treated with 0.6 ml. of freshly prepared 5 per cent. sodium nitrite solution. After 5 minutes 0.6 ml. of 5 per cent. urea solution is added, and the solution is allowed to stand for another 5 minutes until all the bubbles of gas have escaped. The reagent is stable for 6 days if stored in the dark.

criticism in that it is assumed that the pre-digestion process proceeds to completion, although this never occurs in practice and, indeed, is theoretically impossible. This difficulty, however, may be overcome by adding a known amount of aneurin to a separate portion of the sample prior to digestion, and calculating the extent of digestion from a determination of the "added aneurin" content by the prescribed method. Other disadvantages are the incomplete oxidation of aneurin which occurs when the methyl alcohol, sodium hydroxide and potassium ferricyanide are pre-mixed, and the errors of matching the fluorescence visually. The following procedure avoids these difficulties:—The hand-picked and cleaned sample (1 to 2 lb.) is milled finely in a disintegrating mill, and the resulting flour is assayed within 3 days. To two 20-g. portions are added, respectively, 80 ml. of a 0.1 per cent. solution of pepsin in 0.33 per cent. hydrochloric acid; and 79 ml. of the above pepsin-acid solution and 1 ml. of a standard solution containing 100 μ g. of aneurin. After incubation overnight at 37° C., followed by the addition of 2.6 ml. of N sodium hydroxide solution and 100 mg. of taka-diastase, incubation is resumed for 5 hours, portions of each mixture being then centrifuged. To 4 alkali-resistant test-tubes (6 \times 1 in.) are added the following:—To Nos. 1 and 2, 2 ml. of the clear centrifuged liquid containing no added aneurin; to No. 3, 2 ml. of the centrifugate containing the added aneurin; to No. 4, a mixture of 1 ml. each of the standard solution of aneurin and of water. To each tube is then added 2 ml. of methyl alcohol, followed by 0.2 ml. of water and 1 ml. of 30 per cent. sodium hydroxide solution to tube 1, and by 1 ml. of the alkali and 0.2 ml. of a fresh 5 per cent. solution of potassium ferricyanide to tubes Nos. 2, 3 and 4. A stream of nitrogen is passed through the tubes during, and for 1 minute after, these additions, and also for 3 minutes after the subsequent addition (to each) of 25 ml. of redistilled isobutanol which has been saturated with water. The isobutanol layers are then filtered through papers which have previously been extracted for 24 hours with the hot (wet) solvent; the filtrates should not be stored in corked vessels, as fluorescent material may be extracted. The fluorescence is measured in a Hills-Cohen fluorimeter (*Biochem. J.*, 1939, 33, 1966), illumination being provided by a 500-watt mercury arc, which is fitted with two 3-mm. Wood's glass filters separated by a gap of 0.25 inch (which serves as a heat-filter). The fluorescence is measured with a Weston Type "Photronic" cell, fitted with Wratten 2A and 49A filters, and connected to a "Cambridge" short-period galvanometer (resistance, 200 Ω). The solutions are matched in "cells," i.e. thin-walled specimen tubes (2 \times 1 in.) which have no appreciable fluorescence, and the lamp should be standardised between readings against a piece of fluorspar, which is preferable to a solution of quinine

sulphate for this purpose, being more stable. The following readings are made:—(1), Cell No. (1) alone; (2), cell (1) containing 15 ml. of unoxidised extract from tube 1; (3), a second cell, No. (2), alone; (4), cell (2) containing 15 ml. of oxidised extract from tube 2; (5), mixture (4) after addition of 0.15 ml. of oxidised extract from tube 4; (6), cell (5) after a second addition of 0.15 ml. of extract from tube 4; (7), a third cell, No. (3), alone; (8), cell (3) containing 15 ml. of extract from tube 3. The difference values (2)–(1), (4)–(3), (5)–(3), (6)–(3) and (8)–(7) enable the extent of digestion to be calculated and allowed for, and a worked example is given in the original paper. Since the extraction of thiochrome by isobutanol is incomplete, it is important to adhere to the specified conditions so far as possible, so as to maintain a constant partition-coefficient, and in the event of any variations in the quantities taken (e.g. the use of larger aliquot portions owing to the low aneurin-content of a food-stuff) the others must be correspondingly adjusted. When such circumstances necessitate the use of a larger quantity of ferricyanide errors may be introduced, owing to the higher unoxidised blank and the reduction by the ferricyanide of the "blank" fluorescence of the oxidised solutions. If a foodstuff is known to contain no protein-bound aneurin or phosphorylated cocarboxylase (e.g. wholemeal flour), the pre-treatment with pepsin or taka-diastase, respectively, may be omitted, and 0.33 per cent. hydrochloric acid used for the extraction, so long as this procedure does not substantially increase the fluorescence of the blank. The standard deviation for 9 separate assays of the same flour (ranging from 2.05 to 2.25 μ g. of aneurin per aliquot taken) was 0.081 (i.e. approximately 4 per cent. of the mean), and since the fluorescence of the blank rarely exceeds 15 per cent. of that of the oxidised sample and it is unlikely that more than a small fraction of the former is destroyed by oxidation, the accuracy obtainable compares very favourably with that of most biological methods. The aneurin contents of the 78 wheats examined, which represented most available commercial types, were (excluding those of the *Triticum durum* species) 0.54 to 2.60 (average, 1.25); *T. durum* wheats (5 samples), 1.65 to 3.33 (average 2.37); English wheats (33 samples), 0.78 to 1.98 (average, 1.39), I.U. per g. With the exception of *T. durum* the aneurin content bears no consistent relationship to the species, and manuring, soil, climatic conditions or the growing period of the plant fail to explain the variations found; no serious loss of aneurin from wheat occurs after 10 years' storage. The exceptional nature of *T. durum* is evident, even with wheats of this species grown in England, and it indicates that this is a real species difference, which is independent of soil or climate and is possibly related to the fact that the chromosome number of this species differs from that of *T. vulgare*.
J. G.

Bacteriological

Classification and Identification of Bacteria with Special Reference to the Beer Types. J. L. Shimwell. (*J. Inst. Brewing*, 1940, 207-215.)—General principles of classification are discussed, and the criteria available for classification (morphology, motility, orientation, spore formation, cultural characters, physiology, habitat, serological tests, and Gram staining) are reviewed and their limitations pointed out. A proposed key is set out for the determination of the genus of known brewery bacteria, advantage being taken of the fact that only a limited variety of genera occur in that industry. The key is as follows:

1. GRAM POSITIVE.

A. Catalase positive.

- (a) Rods (1) Form spores. Genus: *Clostridium*.
(2) Do not form spores. Genus: *Propionibacterium*.
- (b) Cocci (1) Form cubical packets. Genus: *Sarcina*.
(2) Do not form cubical packets. Genus: *Micrococcus*.

B. Catalase negative.

- (a) Rods (1) Form spores, strictly anaerobic. Genus: *Clostridium*.
(2) Do not form spores, facultative. Genus: *Lactobacillus*.
Species: *L. pastorianum* (and probably many others not yet recorded).
- (b) Cocci (1) Genus: *Streptococcus*.
Species: *S. damnosus* (Clausen)
" var. *pernicius*.
" var. *viscosus*.
S. tetragenus (Walters).

2. GRAM NEGATIVE.

A. Produce acetic acid from alcohol.—Genus: *Acetobacter*.

Species: *A. aceti*, *A. pastorianum*, *A. viscosum*, *A. capsulatum*, etc., etc.

B. Do not produce acetic acid from alcohol.

(a) Motile.

- (1) With peritrichous flagella: Family *Enterobacteriaceae* ["coliform" types, "wort bacteria"].
- (2) With tophotrichous flagella, produce alcoholic fermentation of glucose.
Species: *A. anaerobicum*.

(b) Non-motile, large rods.

- (1) Highly pleomorphic on solid media or in neutral liquid media.
Species: *Flavobacterium proteus* [the common short fat rod of brewers' yeast].
- (2) Not markedly pleomorphic; produce acetylmethylcarbinol.
Genus: *Aerobacter*.

When using this key it must be borne in mind that it is purely provisional, and as time goes on and our knowledge of organisms increases it will have to be amplified to include new types and species. Three plates illustrate *Str. damnosus*, a *micrococcus* species, a *sarcina* species, *L. pastorianus*, *Achromobacter anaerobicum* showing flagella, rosette and club-shaped arrangement, rod forms from pitching yeast and a gelatin streak culture of *L. pastorianus*. D. R. W.

Routine Determination of the Bacterial Content of Paper-board. L. C. Cartwright and S. S. Epstein. (*Paper Trade J.*, 1940, 111, T.A.P.P.I. Sect., 40-51.)—Existing and pending regulations concerning bacteriological standards for paper-board for food containers used in the United States render necessary

routine tests of the following nature:—At least 5 reels are sampled, sheets approximately 10 in. square being taken from 5 layers down and trimmed and cut up under aseptic conditions. Ten g. are weighed and put in a sterile Pyrex flask containing 1 litre of sterile water, and the flask is then plugged with cotton wool and maintained at 35° to 40° C. for 2 to 6 hours. Pulping takes place in a welded, stainless steel churn fitted with a twin-propeller electric mixer; a small hole in the cover serves for the withdrawal (in a sterile pipette) of aliquot portions of the pulped sample, without stopping the mixer or exposing the contents of the churn to contamination. Before use the churn, cover and propellers should be wrapped in paper and sterilised at 15 lb. per sq. in. for

30 minutes, the fixed portions which project inside the churn being wiped with 95 per cent. alcohol and "flamed." In routine work with samples of low bacterial content, steam-sterilisation between each pulping operation is unnecessary, the churn being rinsed with 3 litres of sterile water and the propellers and cover washed in alcohol and "flamed." The course of pulping (which takes 20 to 30 minutes) is followed by withdrawing samples in a sterile 10-ml. pipette, the end of which has been cut off and ground to an internal diameter of approximately 4 mm. This sample is transferred from the pipette to a sterile Petri dish (diameter 100 mm.), a nutrient agar medium being then added and the mixture incubated at 37° C. for 48 hours. Three methods of plating with different proportions of medium are compared statistically, and it is concluded

that there is much less chance of the formation of "spreader"-colonies if the strength of the medium is double the normal value. Thus, ten 5-ml. portions of sample were mixed with 5.5 to 6.5 ml. of a medium containing 0.5, 0.3 and 1.5 per cent. of peptone, beef extract and agar-agar, respectively. If an average of the counts obtained for the 10 plates is taken, the result is more reliable than that obtained by plating the equivalent of 0.1 g. of sample into a single large dish. The bacterial counts recorded for a large number of paper-boards vary from 12 to 273 per g. of sample. J. G.

Formation of Trimethylene Glycol from Glycerol by *Aerobacter*. M. N. Nickelson and C. H. Werkman. (*Enzymologia*, 1940, 8, 252-256.)—The fermentation of glycerol by four strains of *Aerobacter* in a medium of only glycerol and inorganic salts has been found to convert about 45 per cent. of the glycerol into trimethylene glycol. Small amounts of acetyl methyl carbinol and considerable amounts of 2,3-butyleneglycol were also found, but no succinic acid. Braak did not observe the formation of trimethylene glycol with two strains of *Aerobacter aerogenes*, but obtained it under anaerobic conditions with a strain which he named *B. Freundii*. With other strains he found that the fermentation of glycerol was at first vigorous, but was later arrested unless a hydrogen acceptor, such as peptone, was present. The paper sets out in tabular form the percentages of biological products of the author's strains and of Braak's strains in a medium of glycerol and inorganic salts. These include hydrogen, carbon dioxide, formic acid, acetic acid, lactic acid, succinic acid, ethyl alcohol, 2,3-butyleneglycol, and trimethylene glycol. Experimental methods are described.

D. R. W.

Agricultural

Minimum Lethal Dose of Selenium as Sodium Selenite for Horses, Mules, Cattle and Swine. W. T. Miller and K. T. Williams. (*J. Agric. Res.*, 1940, 60, 163-173.)—Selenium in the form of sodium selenite was given in single large doses to horses, mules, cattle and swine, the dose being calculated as mg. of selenium per pound body weight. The quantity was reduced for other animals until the minimum lethal dose was reached. Five horses and 3 mules were used, and the minimum dose for them was about 1.5 mg. per lb. of body weight. For cattle (one calf 5 days old, 2 cows 3 to 4 years and 2 older) the dose was between 4.5 and 5 mg. per lb. of body weight. For swine (7 of 4 to 6 months) the dose was between 6 and 8 mg. per lb. of body weight. The dose varied considerably for the mules, two of which were fat and one was lean, and it seems probable that, at any rate for horses and mules, animals in good condition may be more susceptible to the action of selenium than thin ones. It may be that if the dose were calculated in terms of blood-volume, rather than for body-weight, the difference would not be so pronounced.

D. G. H.

Arsenic in Soils and Waters in the Waiotapu Valley, New Zealand. R. E. R. Grimmett and I. G. McIntosh. (*New Zealand J. Sci. and Techn.*, 1939, 21, 137-160A.)—Sickness and death of cattle, especially dairy cows, on drained swamp land at Reporoa, in the central volcanic plateau of the North Island, have been found to be associated with high arsenic content in soils and waters. The Waiotapu River frequently floods the low-lying parts of the district, leaving a deposit of arsenical mud in the paddocks, and this appears to be largely responsible for the contamination. In one area the draining of the surrounding land caused the activity of hot springs to cease, but in a rain-fed drinking pool in this area there was a crust, about 1 inch thick, of orange-yellow material composed of silica in admixture with about 5 per cent. of orpiment and realgar. Muds from drains and seepage areas in other parts of the district contained arsenic in amounts up to more than 1 per cent. On one farm a spring with a flow of about 1 gall. per minute contained 2.6 grains of arsenic (as As_2O_3) per gall. Samples taken from under the surface of the Waiotapu River at different points showed various amounts of arsenic up to 0.276 per cent., whilst in the topsoils the amount ranged from 0.525 per cent. downwards. The arsenic on the farm lands was found to be in three forms of chemical combination—acid-insoluble, acid-soluble, and water-soluble, the first corresponding with arsenic sulphide and the second with a compound of arsenic with iron. It is pointed out that the area concerned is relatively small, covering about 2000 acres, and is in a thermal district in which the circumstances are very exceptional.

Chemical Effect on Lead Arsenate of Certain Salts which may be present in Soil and Spray Waters. J. M. Ginsburg. (*J. Agric. Res.*, 1940, 60, 199-205.)—To investigate the possibility of soluble arsenic compounds finding their way to the roots of trees or crops where arsenical sprays have been used, some 50 salts generally present in soils or spray waters were tested to find to what extent they could give rise to soluble arsenic when in contact with acid lead arsenate used in the strength of 3 lbs. per 100 gallons of water. This solution was left for 24 hours, with frequent shaking, in contact with various concentrations of the salts. Nitrates, sulphates and acetates proved relatively non-reactive; chlorides, silicates and bicarbonates produced moderate quantities, and salts of carbonates and sulphides large amounts of soluble arsenic. The three phosphates of calcium and monobasic phosphates of sodium and potassium formed inappreciable quantities of soluble arsenic, but the dibasic, and particularly the tribasic phosphates of sodium and potassium produced large amounts. Of a group of salts possessing the property of decomposing lead arsenate, the most highly soluble will form more soluble arsenic than the slightly soluble. Salts with pH values 8 to

11.4 produced, with the two exceptions of sodium and calcium sulphates, more soluble arsenic than salts with lower pH values.

D. G. H.

Manganese Method for Determination of Base Exchange Capacity of Soils and other Materials. C. A. Bower and E. Truog. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 411-413.)—The material is saturated with bivalent manganese by treatment with manganese chloride solution; after displacement, the manganese is determined colorimetrically as permanganic acid. The base exchange capacity of a range of materials tested, *viz.* loams, peat, clays, Bentonite and an artificial zeolite, expressed in milli-equivalents of base per 100 g. of material, was found to be closely similar for manganese and calcium. A 1-g. sample in a 100-ml. stoppered centrifuge tube is shaken vigorously for 5 minutes with 50 ml. of *N* manganous chloride solution. The sides of the tube are washed down with neutral alcohol, and the liquid is centrifuged and poured off. The process is repeated 5 times, which is sufficient to saturate the material with manganese. The excess of manganese chloride is washed out of the material by repeated shaking and centrifuging with 50-ml. portions of neutral 95 per cent. ethyl or methyl alcohol until the washings are free from chloride; 4 washings usually suffice. The exchangeable manganese in the material is now displaced by washing with 5 successive portions of *N* ammonium acetate solution, shaking and centrifuging being used. The washings are combined, and an aliquot portion, containing 0.25 to 0.75 mg. of manganese, is evaporated to dryness. The carbonaceous matter in the residue is destroyed by evaporation with nitric acid. The dry residue is treated with 5 ml. of conc. sulphuric acid, and diluted with about 15 ml. of water; 0.2 g. of sodium periodate is added, and the solution is boiled gently until the colour of permanganic acid appears. The solution is diluted to 90 ml. and kept at 85° to 90° C. for 30 minutes to complete the oxidation of the manganese, after which the solution is cooled, and the manganese is determined colorimetrically.

S. G. C.

Organic

Behaviour of Certain Substituted Allenes towards the Meinel Colour Test. F. B. LaForge and F. Acree, Jr. (*J. Amer. Chem. Soc.*, 1940, 62, 1621-1622.)—Meinel's test (*Ber.*, 1937, 70B, 429) for conjugated double bonds is based upon the treatment of the compound with one molecular equivalent of bromine in methanol solution. The isolated methyl hypobromite addition product is treated with a suspension of silver thiocyanate containing ammonium ferric sulphate. The formation of a red colour was stated to indicate the presence of a conjugated system of double bonds. In the authors' experiments on the reaction of halogens with compounds having a cumulated system of double bonds it was

found that 1-phenyl-1.2-butadiene, 2.3-pentadiene, 1-cyclohexyl-2.3-pentadiene and pyrethron gave a positive Meinel reaction. When compared with myrcene, 1-pentene and styrene dibromide the approximate speed and the intensity of colour were greatest with 1-phenyl-1.2-butadiene and decreased in the order named with the other compounds. As compounds possessing a cumulated system of double bonds also respond to the Meinel test, the method is not specific for the conjugated system.

E. M. P.

Analysis of Rosin Size. E. A. Georgi. (*Paper Trade J.*, 1940, 111, T.A.P.P.I. Sect., 83-84.)—For the determination of free rosin in rosin size, extraction (*e.g.* with absolute or 95 per cent. alcohol) followed by titration of the extract with a standard solution of potassium hydroxide in alcohol is usually advocated. Objections to this are the hydrolysis of saponified rosin, which also contributes to the titration; the extraction of carbonates and bicarbonates; and the difficulty of titrating the highly-coloured extract obtained. The method now suggested is adapted from that used for the titration of wines, and it may also prove suitable for use with other dark-coloured liquids. It depends on the fact that the use of a filter having the same transmission-band as the absorption band of the indicator in its alkaline state, results in the transition from a bright to a dark field at the end-point. The sample therefore, is dissolved in alcohol in a beaker, which is supported over an Eastman's Wratten Filter 52, below which is a heat-absorbing (*e.g.* Corning Aklo) filter, and under the latter an electric light. Titration is then carried out with thymol blue as indicator, the end-point being a change in the intensity of the transmitted light instead of a change in colour. A change in colour may, however, also sometimes be observed because different samples of size vary in their powers of transmission. Free alkali in size is best determined by dissolving 5 to 7 g. of the sample in a beaker in 35 ml. of neutral anhydrous isopropyl alcohol, the alcohol being added a drop at a time at first, with continual stirring, so as to avoid the formation of lumps. An additional 15 ml. of the alcohol are finally added, the beaker is placed in an air-tight container for 4 hours, and the mixture then filtered through a No. 40 Whatman paper (11 to 12.5 cm.), the residue being washed with hot neutral isopropyl alcohol until free from size. The filter-paper is washed with 100 ml. of water (free from carbon dioxide), and the new filtrate is titrated in the usual way with 0.1 *N* hydrochloric acid, with phenolphthalein and methyl orange as indicators.

J. G.

Studies in the Composition of Coal: Extraction of Coal with Quinoline. R. Belcher and R. V. Wheeler. (*J. Chem. Soc.*, 1940, 866-869.)—Suggestions have previously been made (*e.g.* by Vignon, *Compt. rend.*, 1914, 158, 1421) to use quinoline instead of pyridine for the primary extraction of coal in the

determination of resins and hydrocarbons. The present experiments showed that extraction with quinoline is slower than with pyridine and is no more complete. In addition, technical quinoline undergoes a photochemical synthesis on heating, presumably reacting with some of its impurities, and the results may therefore be erroneous.

E. M. P.

Unsaponifiable Matter in Sulphated Oils and Fatty Alcohols. Committee on Oils, Fats and Waxes. No. XIII. Part I. D. Burton and G. F. Robertshaw. (*J. Int. Soc. Leather Trades' Chem.*, 1940, 24, 293-298.)

—It is generally assumed that the unsaponifiable matter in sulphated oils and fatty alcohols is present before sulphation, or has been added after sulphation, and this may lead to mistaken ideas concerning the composition of some sulphated compounds. Thus, higher fatty alcohols or insoluble organic bases may be present as sulphuric esters (which produce special emulsifying properties), or in combination with carboxylic acids or sulphonc compounds, and determination of the saponifiable matter by saponification with 0.5 *N* alcoholic potassium hydroxide solution will not decompose these (unless, possibly, the oil has a high water-content). If, however, the oil is first split (*e.g.* with *N* sulphuric acid) and then saponified, these higher fatty alcohols and insoluble organic bases will be included in the unsaponifiable matter; this method has proved useful for sulphated sperm oil and sulphated fatty alcohols. The method recommended in the S.P.A. Report No. 1 of the Analytical Methods Committee on Determination of Unsaponifiable Matter in Oils and Fats (*ANALYST*, 1933, 58, 203) should be used for oils which are saponified only with difficulty, and contain unsaponifiable matter which is not easily extracted completely with petroleum spirit (*e.g.* sulphated fish, shark and sperm oils); however, although the ethyl ether recommended for extraction removes a wider range of water-soluble substances than petroleum spirit, it has the disadvantage that it also extracts more soap. With one sulphated product made from a raw material containing higher fatty alcohols or their esters it was found that 7 per cent. of the 35 per cent. of unsaponifiable matter was not recovered until the oil had been split with *N* sulphuric acid (Hart's method), and that, even after splitting, 3 per cent. was still not recovered. The procedure was to remove the alcohol, by evaporation, from the saponified soap solution, which was then neutralised to methyl orange with *N* sulphuric acid and boiled for 2 hours with an additional 50 ml. of *N* sulphuric acid; the fatty matter which separated was then extracted with ethyl ether, and a subsequent test showed it to be free from sulphur. These results may be explained by the formation of sulphuric esters from the unsaturated or hydroxyl groups (or both) present in the higher fatty alcohol esters. In the determination of the unsaponifiable matter the sulphated product is saponified, but the sulphuric ester

groups remain intact, and the free alcohols (but not the fatty alcohol sulphuric esters) are extracted by the ether. During the splitting process, however, the alcohols present as sulphuric esters are liberated and are extracted by the ether, although not necessarily completely. The Schindler fractionation method cannot be used for the accurate determination of the unsaponifiable matter in a sulphated oil, partly because of the difficulty of ensuring complete fractionation and partly because decomposition of the sulphuric esters may occur during fractionation. It is therefore suggested that the normal procedure should be followed to the point at which a solution of the β -fraction in alcohol and of α_1 - and α_2 -fractions in petroleum spirit are obtained. The unsaponifiable matter is then determined in the latter by evaporating the solution and applying the S.P.A. method (*loc. cit.*); in the former by neutralising the acetic acid with potassium hydroxide, evaporating to remove the water and alcohol, and after adding 25 ml. of 0.5 *N* alcoholic potassium hydroxide solution, following the S.P.A. method. A solution, in 50 ml. of water, of the residue obtained after evaporation of the soap solution is boiled for 1 hour with 26 ml. of 0.5 *N* hydrochloric acid and 5 g. of sulphuric acid in order to split the sulphuric esters, and the resulting solution is extracted with ether. The ethereal extract is washed with water until free from acid and then evaporated, and the unsaponifiable matter in the residue is determined by the S.P.A. method. The unsaponifiable matters obtained from a sulphated sperm oil were:— α_1 - and α_2 -fractions, 16.7; β -fraction, 4.1; present as sulphuric ester, 13.7; total, 34.5 per cent. The unsaponifiable matter, as determined after splitting with *N* sulphuric acid in the determination of combined SO_3 (Hart's method) was 36.7 per cent., and the difference between this and 34.5 per cent. is mainly accounted for by the presence of unsaponifiable matter in the γ - and δ -fractions, which was not determined. The Wizeff method for this determination, in which the sulphuric acid ester group is removed before the determination of the unsaponifiable matter, is regarded as inaccurate, because the result includes free unsaponifiable matter plus unsaponifiable matter present in combination in sulphuric esters.

J. G.

Oxidation of Drying Oils and Cognate Substances. VI. Properties of the Peroxide, Ketol and Oxido Groupings, including those of some Resins. R. S. Morrell and E. O. Phillips. (*J. Soc. Chem. Ind.*, 1940, 59, 144-148r; *cf. ibid.*, 1939, 58, 159r.)—In continuation of the investigation of the oxidation of drying oils and some resins, the action of hydrogen iodide on substances containing contiguous hydroxyl groups was examined. With polyhydric alcohols the action is variable; with hydrobenzoin, normal; with glycerol, negligible; with ethylene glycol, partial inter-action occurs; with dihydroxy acids, the cis-form reacts the more strongly. A study of the

reactive oxygen values of rosin indicated that nearly 75 per cent. of the calculated polymerised peroxide was formed, and a rosin ester film on drying in air behaves as a drying oil as far as oxidation is concerned. In blonde shellac the R.O. value was found to be 3.31, and a ketone grouping is indicated. A study of enolisation of the ketone grouping has been made, and the discrepancy between the iodine values obtained by the Hübl method (10.2) and Wijs method (87.2) is due to this, the isomeric ketol acids showing 64-99 per cent. enolisation. Whenever a ketol group is present the iodine value will be variable, and its magnitude will depend on the solvent, the temperature, and the symmetry of the molecule. The oxido grouping in oxidoelaidic acid was studied. It is not reduced by hydrogen, forms a hydrobromide, and polymerises at 100° C. to a dimeride. The investigation of the methylation products of oxidised β -elaeostearin has been continued, and earlier results of Morrell and Marks (*J. Soc. Chem. Ind.*, 1931, 50, 27r) have in general been confirmed, with some modifications due to the application of the R.O. values. The esterified product was separated into fractions insoluble and soluble in petroleum spirit, and structural formulae were assigned to them. The reduction of the methyl oxido-methoxy β -elaeostearic acid by hydrogen at atmospheric pressure with a platinum catalyst was unsuccessful as regards the oxido group owing to the very slight fall in R.O. value, accounted for by partial reduction of the methylated ketone grouping. D. G. H.

Effect of Ferric Sulphate in Shortening the Digestion Period in the Kjeldahl Method. F. M. Stubblefield and E. E. Deturk. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 396-399).—The use of the following mixture is claimed to give more rapid digestion than the normal Kjeldahl-Gunning-Arnold process: 25 ml. of conc. sulphuric acid, 0.6 g. of metallic mercury, 10 g. of anhydrous dipotassium phosphate and 6 g. of ferric sulphate.

S. G. C.

Inorganic

Determination of Sodium in Presence of Other Metals. E. C. Elliott. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 416-417).—The method of Caley and Sickman (*J. Amer. Chem. Soc.*, 1930, 52, 4247) employing precipitation with magnesium uranyl acetate reagent, was used. It was confirmed that small amounts of sodium can be determined without interference in presence of beryllium, cerium, lanthanum, neodymium, thallium, thorium, vanadium and zirconium. Silica, if present, should be removed by evaporation with hydrofluoric acid and a little sulphuric acid. Tantalum and niobium form gelatinous precipitates with the reagent and must be separated prior to determination of sodium. S. G. C.

The Fischer Volumetric Method for the Determination of Water. E. G. Almy, W. C. Griffin and S. C. Wilcox. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 392-396).—The Fischer method (*Z. Angew. Chem.*, 1935, 48, 394) consists in titrating the sample with a solution containing methanol, iodine, sulphur dioxide and pyridine; the colour of free iodine appears when all the water has been titrated, indicating the end-point. The present authors propose a potentiometric method for the detection of the end-point to render the process applicable to dark coloured solutions. The titration vessel consists of a flask fitted with a four-holed bakelite stopper accommodating (a) burette nozzle, (b) stem of a paddle for mechanical agitation during titration, (c) platinum wire electrode, and (d) tungsten wire electrode. A thermionic valve type millivoltmeter is required for detection of the potential changes of the electrodes. The reagent is prepared by mixing 203 g. of liquid sulphur dioxide with each 1 kg. of pyridine; to 1359 g. of this mixture, after cooling, are added 1786 g. of methanol followed by 454 g. of iodine, with efficient cooling; the reagent is allowed to "age" for a few days before use. *Method.*—The sample, containing 0.05 to 0.2 g. of water, is mixed with a measured excess of reagent in the titration flask. The liquid is then back-titrated with a solution of water in methanol (5 g. per litre) in order to determine the residual excess of reagent. The end-point is shown by a sudden rise in E.M.F. of the platinum-tungsten couple. A second titration is made to determine the equivalence of the reagent and the alcoholic water solution. The ultimate standardisation of the reagent is carried out by adding a weighed quantity of water to an excess of the reagent and back-titrating with the alcoholic water solution; daily standardisation of the reagent is recommended, owing to a slow change in its water-equivalent. The method is applicable to numerous substances: oils, fats, waxes, monohydric and polyhydric alcohols, glue; insoluble powders may be treated after vigorous agitation with the reagent or prior suspension in anhydrous solvent. Substances which react rapidly with free iodine cannot be dealt with, but if the reaction with iodine is slow a good approximation to the water content can be obtained by carrying out the method rapidly. Inorganic hydroxides interfere by reacting with acids formed in the titration, yielding a salt and water. Acids which react with methanol, liberating water, also interfere, boric acid for example; acetic acid, however, does not interfere, and water in glacial acetic acid may therefore be determined. The nature of the reaction of water with the reagent is obscure (*cf.* Smith, Bryant and Mitchell, *J. Amer. Chem. Soc.*, 1939, 61, 2407). S. G. C.

Modification of the J. Lawrence Smith Method for the Extraction of Alkalis in Rocks. R. E. Stevens. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 413-415).—Barium chloride (1 g.) is substituted for the ammonium

chloride (0.5 g.) customarily used in the sintering mixture. The details of the process are similar to those of the usual J. Lawrence Smith process. The advantages claimed are the elimination of the preliminary slow heating of the charge to remove ammonia, saving about $\frac{1}{2}$ hour, and the automatic removal of sulphate.

S. G. C.

Microchemical

Micro-determination of Bromine in Foodstuffs. W. P. Ford, D. W. Kent-Jones, A. M. Malden and R. C. Spalding. (*J. Soc. Chem. Ind.*, 1940, 59, 177-180r.)—Methods for the determination of bromine in foodstuffs must take into account the presence of relatively large quantities of chlorides, and they fall into 3 categories:—(a) The use of a colorimetric reagent, which reacts with bromine but not with chlorine (*cf.* Seaber, *ANALYST*, 1936, 61, 14). (b) The preferential liberation of bromine by controlled oxidation (*cf.* Francis and Harvey, *Biochem. J.*, 1933, 27, 1545); this is the most direct method, although its accuracy depends on a balance of errors due to the loss of some bromine and the liberation of chlorine (*cf.* Hahn, *Mikrochem.*, 1933, 17, 222). The present authors obtained recoveries of bromine of 85 to 110 per cent. from flour which had been treated with potassium bromate equivalent to 6 parts of bromine per million; this is regarded as satisfactory. (c) Oxidation of bromides to bromate by hypochlorous acid, followed by liberation of iodine from an iodide in acid solution (*cf.* Kolthoff and Yutzy, *Ind. Eng. Chem., Anal. Ed.*, 1937, 9, 75). Advantages are that the accuracy is unaffected by chlorides, and that 1 bromide ion is equivalent to 6 iodine atoms; disadvantages are the large blank correction often involved and the possibility of the formation of chlorates during oxidation. Since the colorimetric method (a) has with very few exceptions hitherto been used for foodstuffs, determinations were carried out by method (b) and a combination of a "protected" ashing process and method (c). When the bromine-content is such that a quantity of sample containing 0.5 g. of dry solids can be used for the determination, the sample is evaporated with 2.5 ml. of 10 per cent. potassium hydroxide solution in a nickel crucible, and the residue is ignited for 40 minutes at 480° to 500° C.; the ash is then moistened with 1.5 ml. of water and dried, and the ignition process is repeated for 20 minutes. If the bromine-content is lower, 2.5 g. of dry solids and 5 ml. of the alkali are used, the crucible being placed in a cold muffle furnace and heated to 300° C. in 30 minutes; the contents are then stirred, and the temperature is raised to 500° C. in 40 minutes. After cooling, addition of 10 ml. of water and drying, ignition is completed at 480° to 500° C. for 1 hour, and the ash is extracted once with 15 ml. and twice with 10 ml. of warm water. The residue from the filtration, together with the filter-paper, are dried with 1 ml. of the alkali and ignited for 45 minutes at 480° to

500° C., the extraction process described above being repeated. Finally, the filtrates are combined and evaporated, and the residues are ignited for 20 minutes at 480° to 500° C. The details given above are important if losses of bromine are to be avoided. In method (c) the ash is extracted 3 times with 0.75 ml. of water "and" (plus?) 20 ml. of alcohol, the alcohol being removed from the combined filtered extracts by placing the flask containing them in a boiling water-bath and sucking air through it. The solution is then heated in a boiling water-bath for 10 minutes under a reflux condenser with 5 ml. of 15 per cent. sodium chloride and 0.75 ml. of 15 per cent. potassium dihydrogen phosphate solutions, and 0.25 ml. of a solution prepared by absorbing 7 g. of chlorine in a solution of 12 g. of potassium hydroxide in 100 ml. of water. Five ml. of water and 1 ml. of 10 per cent. sodium formate solution are added, and the heating is resumed for 10 minutes, the sides of the condenser being again washed down with 5 ml. of water after 5 minutes. The solution is cooled to 22° C. and allowed to stand in the dark with 1 ml. each of 10 per cent. potassium iodide solution, 2.5 per cent. ammonium molybdate solution, and 1.25 ml. of 4 N sulphuric acid. It is then titrated with 0.001 N sodium thio-sulphate solution until pale yellow, 1 ml. of a 2 per cent. solution of Lintner soluble starch being added, and the mixture diluted to 50 ml. in a Nessler cylinder. The titration is completed, comparison being made with a cylinder containing the solution from a titration which had been completed a few minutes previously and kept in the dark in the interval. With a correction for a blank titration, recoveries of bromine of 85 to 106 per cent. from various foodstuffs (and of 97 to 99 per cent. from 60 μ g. of potassium bromide) are obtainable, and only insignificant amounts of chlorate are formed. The necessity for the use of ammonium molybdate to catalyse the reaction between the bromate and iodide ions arises from the fact that the sodium formate used to reduce the excess of hypochlorous acid retards the former reaction. Traces of copper markedly catalyse oxidation of the iodide ion by air, and should be avoided. Any iodine present in the sample also figures in the result, but a study of the literature shows that with normal foods this amounts to only about 2 per cent. of the figures now recorded for the bromine-contents. These results show that the 32 samples of common foodstuffs examined (untreated wheat flour, white and wholemeal breads, tap water, meat, milk, salt, yeast and potatoes) contained 1 to 17 p.p.m. of bromine, and they confirm those of previous workers (which are also tabulated); dried rat's brains contained 23, and dried rat's liver 19 p.p.m. The two methods gave results agreeing well with method (c), being checked in an independent laboratory. Wheat flours guaranteed to be free from chemical treatment contained 2.4 to 7.7, and white breads 1.6 to 5.4 p.p.m. "Treatment" of flour with potassium bromate containing bromine equivalent to 3.6 p.p.m.

raised the bromine-content of the bread by only about 2.5 p.p.m., so that it is not possible to ascertain from the bromine-content if a flour has been so treated. J. G.

Microchemical Determination of the Reducing Power of Cellulose, Oxycellulose and Hydrocellulose (Micro-Copper, Micro-Ferricyanide and Micro-Iodine Numbers). E. Geiger and G. Müller. (*Helv. Chim. Acta*, 1940, 23, 820-826).—The relative merits of the 3 determinations are compared, and the results obtained on the macro-scale with rayon and oxycellulose (oxidised rayon) are tabulated. The values found, respectively, were:—Copper number, 12.4, 54; iodine value, 10.0, 19.0; ferricyanide number, 56.9, 133.6. The ratios oxycellulose/regenerated cellulose for the 3 methods were, respectively, 4.37, 1.90 and 2.35. The copper number is considered to give the most useful and consistent results, although it is influenced appreciably by the experimental conditions (e.g. concentration, alkalinity and quantity of sample used), and these must be observed closely in carrying out the micro-modification of the method. The basis of this is the determination of ferrous iron colorimetrically, instead of by titration with potassium permanganate solution. *Micro-copper number* (cf. Knecht and Thompson, *J. Soc. Dyers & Colourists*, 1920, 36, 255).—The sample (0.1 to 10 mg., corresponding with a copper number of 10 to 0.1 g. of copper per 100 g. of sample) is sealed in a tube (length 3 cm., diameter 2.5 mm.) with 0.04 ml. of Braidley solution (i.e. a mixture of 5 ml. of a 10 per cent. solution of crystalline copper sulphate, and 95 ml. of a solution containing 50 g. of sodium carbonate per litre). The tube is immersed for 3 hours in a boiling water-bath, and then cooled and opened, the sample is removed with a fine glass hook and the liquid is filtered in a sintered glass micro-crucible. The residue is washed first with the Braidley carbonate-bicarbonate solution, then with 1 drop of 2 N sodium carbonate solution and finally 3 times with 3 drops of cold water. The filtrate is rejected, and the cuprous oxide in the residue is dissolved in 1 drop of a solution containing 5 per cent. of ferric sulphate and 20 per cent. of sulphuric acid; 3 drops of water, 1 drop of 2 N sulphuric acid and three 3-drop portions of water are used, in succession, for washing. Then the ferrous salts in the resulting filtrate are determined by adding 3 drops of a 1 per cent. solution of α,α' -dipyridyl or (preferably, since it is more sensitive and cheaper) of *o*-phenanthroline hydrochloride, followed by 1 drop of 10 per cent. ammonium acetate solution, a deep red or orange-red colour, respectively, developing in about 10 minutes. Three drops of a 1 per cent. solution of sodium fluoride and sufficient water to bring the total volume to 10 ml. are then added, and the colour is matched against that produced in a similar way from a 0.0152 per cent. solution of ferrous sulphate, which has been standardised with 0.04 N potassium permanganate solution, a volume equivalent

to 20 to 80% of ferrous sulphate being diluted to 10 ml. for the purpose; allowance is made for a blank determination. Beer's Law applies for concentrations up to 0.2 mg. per 100 ml., but a calibration curve is necessary with higher concentrations. *Micro-iodine value* (cf. Bergmann and Machemer, *Ber.*, 1930, 63, 316, 2304).—The sample (0.25 to 25 mg., corresponding with an iodine value of 20 to 0.2 ml. of 0.1 N iodine solution per g. of sample) is treated for 90 minutes with 1 ml. each of 0.25 N sodium hydroxide and 0.005 N iodine solutions. The unused iodine is then back-titrated with 0.005 N sodium thiosulphate solution in presence of 0.3 ml. of N sulphuric acid. *Micro-ferricyanide number* (cf. Freiburger *Melliand's Textilber.* 1930, 11, 127).—The sample (0.5 to 50 mg., corresponding with a ferricyanide number of 200 to 2 g. of potassium ferricyanide per 100 g. of sample) is treated with 1 ml. each of 0.005 N potassium ferricyanide and 10 per cent. sodium hydroxide solutions for 10 minutes in a boiling water-bath. The mixture is rapidly cooled, and 0.5 ml. of 30 per cent. acetic acid, 1 ml. of a solution containing 50 g. of zinc sulphate and 10 g. of sodium chloride per litre and a crystal of potassium iodide are added in succession. The unused ferricyanide is then back-titrated with 0.005 N sodium thiosulphate solution, allowance being made for a blank determination. Results are tabulated for cotton linters and for viscose rayon alone and containing oxycellulose or hydrocellulose. Slight deviations between duplicate results obtained for the same sample are attributed to sampling errors. The silver nitrate method for the determination of reducing power (cf. Götze, *id.*, 1927, 8, 624, 696) is considered unreliable. J. G.

Thorium Nitrate Micro-titration of Fluorine in Aqueous and Alcoholic Systems. J. W. Hammond and W. H. MacIntire. (*J. Assoc. Off. Agr. Chem.*, 1940, 23, 398-404).—A brief review of the difficulties encountered in the determination of small amounts of fluorine by titration with thorium nitrate solutions is given, and also an account of experiments to determine (a) the influence of fluorine concentration upon the accuracy of the titration in buffered aqueous and 48 per cent. alcoholic systems and in corresponding systems in which the pH is adjusted by means of 0.05 N hydrochloric acid; (b) the conditions under which the value of the thorium nitrate solution follows the stoichiometric equation, and those under which its value is empirical and determinable only by standardisation against standard fluorine solutions. In this work precautions were taken to prevent coatings of colloidal silica in the distillation flasks and to maintain the distillation temperature at 135°C. Two drops of a 0.05 per cent. aqueous solution of sodium alizarin sulphonate indicator per 10 ml. were used in each titration. The aqueous and alcoholic systems were brought to pH 3.0 \pm 0.2, and one series of each type was neutralised and

adjusted to that pH with 1 ml. of 0.05 *N* hydrochloric acid per 20 ml., and another series of each type was similarly adjusted with 1 ml. of buffer solution containing 9.448 g. of monochloroacetic acid and 2 g. of sodium hydroxide per 100 ml. The thorium nitrate solutions were 0.0175 *N* and 0.00175 *N* for respective titrations of fluorine concentrations of mg.-range and γ -range per 10 ml. The two solutions were standardised against ammonium oxalate and against rock phosphate No. 56 of the Bureau of Standards. The fluorine concentration ranges used were 2 to 50 γ per 10 ml. (γ -range) and 0.2 to 5 mg. per 10 to 20 ml. (mg.-range). In alcoholic systems, in the γ -range moderately good agreement was obtained between the acid-adjusted solution and the buffered solution, the buffered solutions giving the higher value, which was also higher than the true value. In aqueous systems, in the γ -range the results in both series of solutions were greatly in excess of the true value, and in the buffered solution again in excess of those in the acid-adjusted solution. In the mg.-range good agreement with the true value was obtained in all the series—aqueous, alcoholic, acid-adjusted and buffered solutions. Blank determinations gave higher results with the buffered solutions in all series in both ranges, and in the γ -range magnified the error in the quantity of fluorine indicated by the titration, particularly in the aqueous system. The results show that micro quantities of fluorine cannot be determined in aqueous systems if the normality value of the thorium nitrate solution is used. For such quantities the value of the solution must be determined empirically against corresponding known quantities of fluorine and for the specific solvent, definite volume, and identical quantity of indicator. When titrations are made in alcoholic solution, however, application of the stoichiometric value of the thorium nitrate solution will give accurate results in a solution of adjusted pH without inclusion of a buffer solution.

A. O. J.

Photographic Silver-gelatin Paper as a Reagent in Spot Analysis. G. Schwarz. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 369–372.)—Photographic paper developed to complete blackness reflects more brightly after being dipped in hot water. This brightening is prevented by all mercaptans and seleno alcohols, by certain heterocyclic substances containing imino groups, by salts of the nobler metals, by iodides, and by substances which easily split off selenium or tellurium, and the reaction is applied as a spot test for these substances. *Detail.*—Silver chloride or slightly sensitive silver chloride-bromide glossy paper (Velox, Ridax, etc.) is suitable, but any new paper should be tested before use. A suitable paper is fully exposed by artificial light or daylight and fully developed, without exclusion of light, the usual developer and fixing baths and considerable excess of developer being used. The paper is washed for 1 hour in tap water, then in frequent changes of distilled water, and dried and cut into suitable sizes for testing; it will keep indefinitely. To test a reagent paper, a piece is dipped halfway into water at 70°–80° C.; in about 3–5 seconds a very distinct decrease of the blackening must be visible. A small drop of the test solution is placed upon the dry reagent paper and allowed to evaporate or, alternatively, the excess is removed with filter-paper after 5 minutes, but this gives a less sensitive reaction. The strip of paper is then dipped into hot water at 70°–90° C.; the reaction is positive if the spot upon which the drop was placed is black against a gray background. The used paper may be dried and kept as a permanent record. Forty-two organic compounds were tested, and the limits of identification in acid and alkaline media are given. In addition, 14 inorganic compounds were tested. For some substances the test was sensitive to a limiting concentration of 1:100,000.

J. W. M.

Reviews

ELECTROCHEMISTRY AND ELECTROCHEMICAL ANALYSIS. VOL. II, GRAVIMETRIC ELECTROLYTIC ANALYSIS AND ELECTROLYTIC MARSH TESTS. By H. J. S. SAND. Pp. ix + 149. London: Blackie & Son, Ltd. Price 5s.

Dr. Sand's name has been for so long associated with electrochemistry and electrochemical analysis that he was the natural choice for the authorship of a modern book, in English, to succeed the classic works of Edgar F. Smith and Classen. The preface to Vol. I announced the completion of the work in a second volume, but it has since been found impossible to include all that was intended, and Potentiometric and Conductimetric Titrations, Moisture determinations by means of capacitance measurement and the electrical measurement of pH have been left for the third volume, to be published when circumstances permit.

In this, the second volume, there are chapters on Apparatus for gravimetric electrolytic analysis, Technique of electro-analytical deposition, Quantitative deposition and separation of individual metals, Separations, Application to the analysis of industrial alloys, Internal electrolysis, Electrolytic micro-analysis, and Electrolytic Marsh tests. The use of the auxiliary electrode is described, as are the various measuring devices for the control of cathode potential—an essential to the separation of metals by graded potential. The usefulness of the section on Internal electrolysis, a method which Dr. Sand and his collaborators at the Sir John Cass Institute have done so much to develop during the last decade, is increased by the inclusion of details of a number of actual separations. In the chapter on quantitative deposition and separation of individual metals the gap existing between theory and practice is revealed. Suitable conditions for the deposition of the commoner metals are given in the form of actual examples; indeed, they are referred to as "prescriptions." Complex ion formation, metal overvoltage, initial concentration of the electrolyte, efficiency of stirring, type of electrode, volume of solution, and even the shape of the beaker, are among the factors which control the separations, so that a measure of empiricism is inevitable. In general, examples are given of depositions from various media, *e.g.* silver from nitrate, ammoniacal, cyanide, and acetate solutions.

The section devoted to the separation of metals is not so comprehensive as might have been expected, but there is a chapter dealing with the analysis of yellow metal alloys, white metal alloys, nickel bronzes and aluminium alloys. The section on micro-analysis contains a list of references to original papers: Pregl's method is described, and also Lindsey and Sand's method for micro-deposition under controlled potential. Descriptions of this and many other developments mentioned in the book have already appeared in *THE ANALYST*.

In the preface, Dr. Sand craves pardon for having given any undue prominence to those developments with which he has been personally associated. This characteristic modesty of the author is, in a sense, reflected in the book. The reviewer would have preferred to see Dr. Sand's life work perpetuated, not in three small pocket books, but in one complete volume which could take its place on the shelf with Treadwell and Sutton and Hillebrand and other classics. Not only would this have done more adequate justice to the author, but it would the more readily have achieved his aim, "to contribute to the more extensive employment of electrolytic methods." It is probably true that these are too seldom used, and while the price of platinum is no doubt an adverse factor, lack of appreciation of the elegance and speed of the methods is also responsible. Dr. Sand's book should go far to remedy this state of affairs, but not so far as it might have gone had there not been a tendency to divide its appeal between the student and the practising analyst.

R. C. CHIRNSIDE

PROPERTIES OF ORDINARY WATER SUBSTANCE. By N. E. DORSEY. Pp. xxiv + 673. New York: The Reinhold Publishing Corporation; London: Chapman & Hall, Ltd. 1940. Price 90s. net.

This monumental compilation of physical data concerning steam, water and the ices having the ordinary isotopic composition was begun under the auspices of a committee of the National Bureau of Standards. In addition to relevant data from the International Critical Tables, revised and supplemented in the light of work published as late as 1937, there is much other information about water, including references to its synthesis and decomposition, but chemical reactions with other substances and solubilities in it of materials other than certain gases are omitted.

Those who occasionally consult the International Critical Tables not infrequently encounter difficulty in determining the precise meaning of the data, but this objection is largely absent from the book under review as a result of the explanations in the tables. It is, however, unfortunate that some device is not uniformly employed to distinguish explanatory matter associated with the tables from the text, which is frequently inserted as a few lines between tables and is then, without notice, continued somewhere on a subsequent page which has to be discovered by trial and error.

Notwithstanding this defect, which could be avoided easily in a future edition, the author is to be congratulated on producing a valuable work of reference.

J. G. A. GRIFFITHS

FORENSIC CHEMISTRY. By H. T. F. RHODES. Pp. viii + 214. London: Chapman & Hall, Ltd. 1940. Price 12s. 6d. net.

This volume is a compilation of information widely diffused throughout criminological literature and a description in brief of the work of a forensic chemical laboratory. Part I deals with personal identification by means of skin prints, occupational dust, blood grouping and seminal stains, and the remainder of the volume gives details of the examination of stains, firearms, documents, inks, paper and subsidiary materials such as sealing wax and adhesives. Methods adopted in the investigation of banknotes and coins are described. The last chapter deals with the isolation and identification of toxic agents, including abortifacients, acids, alkalis, metallic salts, volatile poisons, alkaloids, synthetic drugs and toxic gases. It will thus be evident that much has been crowded into a relatively small space, but the author's excellent literary style has prevented undue compression and sacrifice of lucidity.

The practice of forensic chemistry necessitates the highest degree of accuracy possible, but, unfortunately, much of the subject matter of this volume fails lamentably in this respect. In several instances the only tests given for a particular substance are comprehensive in character and yield the same result with various other compounds, notwithstanding the author's statement on p. 41 that "No test of a general kind can supply precise information."

The different subjects have received very unequal treatment; thus some 12 pages are devoted to the development of skin prints, 6 to the constitution of tannic acid, iron tannates and haematoxylin, and less than 2 to the toxic gas carbon monoxide, since according to the text "this is the only gas which has much forensic importance." To the reviewer the section on "spot" tests for metals appeared curiously familiar, and a brief comparison showed that these had been abstracted almost *in toto*, with slight transposition and without acknowledgment, from a well-known trade publication. Unfortunately during the process a few errors have crept in, whilst the table of micro-reactions for the common metals given on p. 21 perpetuates two errors which occur in the original source and should have been eliminated. Adoption of some of the methods described for the examination of

dusts would lead to the loss of valuable material, and some of the statements relating to this subject are both contradictory and inaccurate.

The section dealing with the isolation of alkaloids and other substances in a pure state from viscera conveys the impression that this is a simple matter, and the smaller details upon which success so largely depends, and which are acquired only by extensive experience, are missing.

The volume concludes with a list of references arranged in alphabetical order of the authors' names, a name index and an accurate, though incomplete, subject index. It is greatly to be regretted that this handbook, which might have been a valuable production, is so seriously marred by errors of various kinds; a thorough revision will be necessary before it can be recommended.

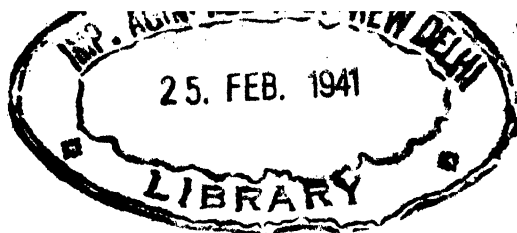
T. J. WARD

CHAMBERS'S TECHNICAL DICTIONARY. Edited by C. F. TWEENEY and L. E. C. HUGHES. Pp. viii + 957. London: W. R. Chambers, Ltd. 1940. Price 15s. net.

This dictionary supplies a distinct want, for every science, industry and trade has its words of specialised meaning, many of which have crept into everyday language and are often only imperfectly understood by those who use them. The scope of the work is very wide, for it covers all the pure and applied sciences, medicine and psychoanalysis, construction, including buildings, bridges and ships, all the principal manufacturing industries including raw materials, processes and machinery, crafts and so forth. Authorities in each subject have contributed definitions of terms used in their respective fields, and most of these can be grasped by the layman. The chemical definitions, however, for which Dr. C. J. W. Hooper, Dr. R. G. Israel and Mr. I. Singleton are responsible, are often of the nature of the definitions in a chemical dictionary, since they give structural formulae, melting-points and reactions, and could not be understood by those who have no chemical knowledge. It is difficult, however, to see how a ketone, for instance, could be defined in such a way as to be understood by anyone who knows nothing of organic chemistry; the problem seems insoluble.

A useful feature is the introduction of registered trade names, such as Cellomoid, Pilanco, Maxweld, Pressspahn, which are spelt with capitals to distinguish them from ordinary trade names. The pronunciation of all the words defined is clearly indicated, and the book is printed in clear type, so that there is no difficulty in finding any given entry. It concludes with a classified bibliography of standard works on the principal sciences, industries and trades. The editors, contributors and publishers may be congratulated on the production of a very valuable work, which will be kept in constant use by those who have it.

EDITOR



THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

The Examination of Lard

By R. W. SUTTON, B.Sc., F.I.C., A. BARRACLOUGH, B.Sc., F.I.C.,
R. MALLINDER, B.Sc., F.I.C., AND O. HITCHEN, B.Sc., F.I.C.

(Read at the Meeting, December 6, 1939)

THE term "lard" is now applied to the fat from any part of the pig, and only relatively small quantities of the product originally limited to the leaf fat are still sold as "butchers' lard" or "home-rendered lard." There is therefore a much wider variation than formerly in the composition of lard, and this involves greater possibilities of adulteration. A considerable amount of fat produced by fat-renderers from the waste from butchers' shops, pigs' bones, etc., is refined and sold, either alone or in admixture with lard. Beef products may readily occur in these mixtures, and during the last two years we have met with numerous samples undoubtedly containing beef fat—in one instance as much as 50 per cent. The difficulty of detecting adulteration is still further increased by the availability of hydrogenated oils of almost any desired consistence. Hence, routine tests such as the determination of the iodine value, refractive index, specific gravity, and melting and solidifying points are no longer sufficient criteria of genuine lard. Other tests for particular types of adulterants must be made, and we recommend in particular that the Bömer value of all samples should be determined.

It has been shown that extraordinary variations may occur in the major component fatty acids of lard (palmitic, stearic, oleic and linolic acids) according to the age of the pig^{1,2} and the character of the ration, and such variations, of course, result in some highly abnormal figures for the analytical constants usually determined.

Thus, König and Schluckebier¹ observed a rise in the melting-point of the body fat and a fall in its iodine value as pigs grow older. Ellis and Hankins² confirmed these results and concluded that ingested fat is deposited to some extent with only slight modification of its glyceride structure. In young pigs, ingested fat forms a high proportion of the body fat; in older animals, an increasing quantity of the body fat is derived by synthesis from carbohydrates and protein in the feed, and this fat is usually of a harder type. Callow³ reports a correlation between the iodine value and the rate of deposition of the fat.

Ellis and Isbell⁴ found that, with a surplus of fatty food, ingested fat tends to be deposited in the body fat. Their analytical results related to samples of back fat from hogs on different rations, ranging from brewers' rice with protein supplement (which resulted in a very hard lard particularly low in linolic acid)

to soya beans and peanuts (which resulted in very oily carcasses). Some of their results are given in Table I.

TABLE I

| | Iodine value | Satd. acids Per Cent. | Oleic acid Per Cent. | Linolic acid Per Cent. |
|--|-----------------|-----------------------------|----------------------------|------------------------------|
| 1. Brewers' rice and protein supplement | 54.7 | 37.5 | 56.1 | 1.9 |
| 2. Corn with protein supplement .. | 60.8 | 37.4 | 49.5 | 8.2 |
| 3. Peanut meal 1 part, corn meal 2 parts | 72.6 | 30.2 | 52.6 | 13.0 |
| 4. Soya beans + 2.5 per cent. of corn | 78.3 | 30.8 | 44.1 | 20.0 |
| 5. Peanuts | 89.6 | 19.5 | 55.0 | 20.3 |
| 6. Soya beans | 93.2 | 26.0 | 39.1 | 30.6 |

In a later paper Ellis and Zeller⁵ refer to lard with iodine values of 30 and 100. These were the result of different rations—copra cake on the one hand, and peanuts or soya beans on the other.

Battacharya and Hilditch⁶ also found that the presence of arachis oil in the diet of pigs led to an increase of unsaturation in the stored fat and to an increase in the proportion of linolic acid in the unsaturated acids.

IODINE VALUE.—Various limits have been proposed for the iodine values of average commercial lards. Our results suggest that in some instances the upper limit should be increased. Whereas years ago an iodine value of 66 was rarely exceeded, figures as high as 69 and 70 are by no means uncommon to-day. Table II gives the values obtained on 113 samples examined since 1933. Many lards with high iodine values have been fully investigated but without yielding evidence of adulteration.

TABLE II

IODINE VALUES OF LARDS

| | | | | | | |
|----------------|----|-----------|-----------|-----------|-----------|------------|
| Iodine value | .. | Below 57 | 57.0-58.9 | 59.0-60.9 | 61.0-62.9 | 63.0-64.9 |
| No. of samples | .. | 2 | 3 | 13 | 16 | 24 |
| Iodine value | .. | 65.0-66.9 | 67.0-68.9 | 69.0-70.9 | 71.0-72.9 | Above 72.9 |
| No. of samples | .. | 18 | 21 | 8 | 5 | 3 |

Of the 113 samples examined, 108 (96 per cent.) fall within the range of 57 to 73.

REFRACTIVE INDEX.—Although the figures recorded by Ellis and Isbell^{3,4} range from 1.4582 to 1.4636 at 40° C. (equiv. to Zeiss butyrometer readings of 48.3 to 56.5), most commercial samples have values falling within a much narrower range. Our results on 105 samples examined since 1933 are given in Table III. The figures tend to be concentrated between 50 and 52.

TABLE III

ZEISS BUTYROMETER READINGS OF LARDS

| | | | | | | |
|----------------|----|------------|-----------|-----------|-----------|-----------|
| Zeiss reading | .. | Below 49.7 | 49.7-50.0 | 50.1-50.4 | 50.5-50.8 | 50.9-51.2 |
| No. of samples | .. | 0 | 4 | 9 | 20 | 32 |
| Zeiss reading | .. | 51.3-51.6 | 51.7-52.0 | 52.1-52.4 | 52.5-52.8 | 52.9-53.2 |
| No. of samples | .. | 21 | 16 | 1 | 1 | 1 |

THE BÖMER VALUE.—The glycerides which separate from an ethereal solution of lard, etc., include most of the higher fully saturated and probably a proportion of the mono-unsaturated glycerides. Since lard normally contains only about 5 per cent. of fully saturated glycerides,⁷ whereas beef fat contains about 17 per cent.,⁸ considerable differences in the amounts of deposits are to be expected. The amount obtained from mixtures, however, varies considerably with the particular samples of lard and beef fat, and no test (including that of Stock⁹) is capable of giving satisfactory quantitative results.

Emery,^{10,11} examining the m.p. of the crystals, concluded that "when the melting-point of the separating glycerides is below 63.4° C. the presence of beef

fat should be suspected, while a melting-point of 63° C. or below can be regarded as positive evidence that the sample is not pure lard." His procedure is substantially reproduced in the British Pharmacopoeia, 1932.

Bömer¹² investigated the saturated glycerides of lard by repeated fractional precipitation and crystallisation from ether. He concluded that tri-stearin was not present and that the glycerides consisted of palmito-distearin (m.p. 68.5° C.) and stearo-dipalmitin (m.p. 58.2° C.). He also found that the palmito-distearin derived from lard differed in m.p. and crystalline form from the palmito-distearin derived from mutton tallow, and concluded that the former was α -palmito-distearin and that the latter contained the palmitic residue in the β -position.

These were based on comparison with synthetic glycerides, prepared by methods which were later discredited, and they were superseded by configurations given by Amberger and Wieschahn.¹³ The palmito-distearin (m.p. 68° C.) separating from lard was therefore accepted as the β -palmito- $\alpha\alpha'$ -distearin, and the glyceride of beef tallow (m.p. 63.3° C.) was thought to be the α -palmito- $\alpha'\beta$ -distearin. Whilst the palmito-distearin with m.p. 68° C. is doubtless β -palmito-distearin, the compound with m.p. 63.3° C. is not necessarily α -palmito-distearin, but quite possibly β -palmito-distearin containing a little myristo-palmito-stearin or dipalmito-stearin. Many slightly impure palmito-stearins melt at 63–65° C.¹⁴

Bömer¹⁵ subsequently extended the test to the fatty acids of the separated glycerides. He found that the difference between the m.p. of the glycerides and of the fatty acids was about 5.2° C., and in some lards reached 6.9° C. With beef and mutton fats the differences ranged from 0.1° to 2.6° C. He concluded that with glycerides melting between 60° and 61° C. the presence of beef fat is indicated when the difference between the m.p. of the glycerides and of the fatty acids is less than 5.0° C., and with glycerides melting between 65° and 68.5° C., when the difference is less than 3° C.

Sprinkmeyer and Diedrichs¹⁶ applied the method to numerous samples of genuine lard, beef and mutton fats, and also to mixtures. With lards the difference between the m.p. of the least soluble glycerides and their constituent fatty acids ranged from 4.4 to 7.4° C.; with beef and mutton fats the difference ranged from 0.8° to 1.2° C. They suggested the factor $Mg + 2d$, which is the m.p. of the glycerides plus twice the difference, and found this value for lard to range from 73.1 to 76.5 and for beef and mutton fats from 65.2 to 67.3. With mixtures containing 5 to 10 per cent. of beef or mutton fat the value $Mg + 2d$ was always below 72 and usually below 70. The presence of hydrogenated oils or fats in lard affected the factor $Mg + 2d$ to the same extent as the presence of beef fat. Bömer¹⁷ found that with lards the values for $Mg + 2d$ always exceeded 71, whilst the values with beef fat, mutton fat and horse and goat fats were well below this figure.

Jesser¹⁸ obtained results for $Mg + 2d$ ranging from 73.3 to 76.8. Prescher¹⁹ examined 58 fats of known character by the method of Bömer, and also using the difference value formulated by Polenske.^{20,21} Of 25 samples of adulterated lard, in only three, containing respectively 10, 20 and 30 per cent. of beef tallow, could adulteration be detected by the Polenske method; the others, some containing as much as 15 per cent. of beef tallow, gave negative results. The Bömer method failed with only 2 fats—in which 5 and 10 per cent. of beef tallow were present. Eighteen samples of pure lard were passed as satisfactory by the Bömer method, whilst the Polenske procedure gave false indications of adulteration with two.

Vitoux and Muttelet,^{22,23} used acetone for the first crystallisation of the glycerides, and suggested the use of a factor equivalent to $Mg + d$ (in place of $Mg + 2d$); the introduction of this variation does not appear to offer any advantage.

Details of the test as elaborated by Bömer and his co-workers are published in "*Einheitliche Untersuchungen für die Fett- und Wachsindustrie*," issued by the German Commission for Fat Analysis.

In 1920 Kerr²⁴ published details of a test in which only 5 g. of fat were dissolved

in acetone, and the crystallisation was allowed to take place at 30° C. The melting-points of the separating glycerides and of their constituent fatty acids were determined, and the factor $Mg + 2d$ was calculated. Using these conditions Kerr proposed 73° C. as the minimum figure for genuine lards. This test was described in detail in the "*Official and Tentative Methods of Analysis of the Association of Official Agricultural Chemists*," and is still included as a "tentative" method in the 4th Edition, published in 1935. It is stated that "when the melting-point of the glycerides obtained by this method is below 63.6° C. the presence of beef fat or other fat containing tristearin should be suspected, and a melting-point of 63.2° C. or lower is evidence that the sample is not pure lard." In the errata to this edition²⁶ is the direction that "if the melting-point of the glycerides + twice the difference between the melting-point of the glycerides and the melting-point of the fatty acids is less than 73° C. the lard is regarded as adulterated."*

In most of our work we have used the procedure specified by the A.O.A.C., and we have found no reason to dispute the standards adopted. In the examination of 15 genuine lards, most of which had been purchased from small pork-butchers' shops as "home-rendered," the lowest figure for the m.p. of the separating glycerides was 64.5° C.; the "difference" figures ranged from 4.2 to 6.1° C., and the factor $Mg + 2d$ from 73.4 to 76.9. Various specimens from a pig's carcase were obtained from a local pork butcher and the fat was rendered in the laboratory. Our results, given in Table IV, also include the figures for two commercial articles described as "Refined lard—prepared from commercial hog fat" and "ham bone fat."

TABLE IV

| Source of fat | Iodine value | Butyro-ref. reading 40° C. | Glycerides from acetone (A.O.A.C.) | | | |
|-----------------|--------------|----------------------------|------------------------------------|-------------------------|-----------|-----------|
| | | | M.p. °C. | M.p. of fatty acids °C. | Diff. °C. | $Mg + 2d$ |
| Back | 61.8 | 50.4 | 64.7 | 59.6 | 5.1 | 74.9 |
| Leaf | 56.4 | 49.5 | 64.9 | 60.6 | 4.3 | 73.5 |
| Midrum .. . | 50.2 | 48.4 | 64.4 | 58.3 | 6.1 | 76.6 |
| *Collar .. . | 70.8 | 51.7 | 65.3 | 60.2 | 5.1 | 75.5 |
| Leg bones .. | 67.1 | 51.2 | 64.7 | 60.0 | 4.7 | 74.1 |
| H-bones .. . | 63.9 | 51.9 | 64.8 | 60.4 | 4.4 | 73.6 |
| Refined lard .. | — | — | 65.0 | 61.0 | 4.0 | 73.0 |
| Ham bone fat .. | — | — | 64.7 | 59.6 | 5.1 | 74.9 |

*Very soft consistence. Ten g. required to give glyceride separation.

Our results on various beef products and some hydrogenated oils and fats, given in Table V, are in agreement with the published work. It will be seen that hydrogenated products usually, but not invariably, cause a lowering of the Bömer value. The hydrogenated sesame oil had a Bömer value of 74.5, but it should be noted that the m.p. of the glycerides was as high as 68.7° C. In particularly hard fats—beef suet and a hydrogenated cotton-seed oil with iodine value 42—the m.p. of the fatty acids was slightly higher than that of the corresponding glycerides.

In attempts to obtain approximately quantitative results by the Bömer test standard mixtures of genuine lard and beef fat were made and the Bömer values were determined. Table VI contains our results. A regular gradation in the figures is to be observed, and this is shown more clearly by the continuous line in Fig. 1.

With a harder type of fat, such as beef suet, a greater variation in the Bömer figures was observed. The results were somewhat irregular for mixtures containing more than 40 per cent. of suet. A heavily hydrogenated cotton-seed oil (iodine value 42) in admixture with lard also caused a rapid fall in the Bömer values. The results were uniform up to about 30 per cent. admixture and thereafter irregular.

* For a recent report on the Bömer method by a Committee of the American Chemical Society see ANALYST, 1940, 65, 508.

In Fig. 1 the central discontinuous line refers to mixtures of lard and beef suet, and the lowest curve to the mixtures of lard and hydrogenated cotton-seed oil. Two mixtures of lard with 20 and 40 per cent. of beef dripping gave Bömer figures similar to those obtained from the corresponding mixtures with beef suet.

TABLE V

Glycerides from acetone (A.O.A.C.)

| Source of fat | Iodine value | M.p. °C. | M.p. of fatty acids °C. | Diff. °C. | Mg + 2d |
|-----------------------------------|--------------|----------|-------------------------|-----------|---------|
| Beef fat | — | 61.6 | 60.9 | 0.7 | 63.0 |
| „ dripping | — | 60.6 | 59.8 | 0.8 | 62.2 |
| „ suet | — | 61.4 | 61.6 | —0.2 | — |
| Edible beef fat | — | 61.0 | 60.6 | 0.4 | 61.8 |
| Crude tallow | — | 61.5 | 61.5 | 0 | 61.5 |
| Beef tallow, No. 7 | — | 60.5 | 60.5 | 0 | 60.5 |
| „ „ „ 8 | — | 60.7 | 60.6 | 0.1 | 60.9 |
| * „ marrow fat | — | 59.2 | 56.0 | 3.2 | 65.6 |
| <i>Hydrogenated oils and fats</i> | | | | | |
| Ground nut | 73.5 | 66.2 | 63.2 | 3.0 | 72.2 |
| Lard | 57.4 | 61.8 | 57.6 | 4.2 | 70.2 |
| Palm | 40 | 60.4 | 54.6 | 5.8 | 72.0 |
| Pig fat | 43 | 61.9 | 57.9 | 4.0 | 69.9 |
| Sesamé | 56 | 68.7 | 65.8 | 2.9 | 74.5 |
| Cotton-seed | 42 | 59.8 | 60.0 | —0.2 | — |
| „ | 68 | 54.5 | 49.7 | 4.8 | 64.1 |
| Whale | | | 52.7 | 4.2 | 65.3 |

Very soft fat. Fifteen g. used and glycerides separated only after about 8 days.

TABLE VI

Melting-points, °C.

| | Glycerides | Fatty acids | Diff. °C. | Mg + 2d |
|--------------------------------|------------|-------------|-----------|---------|
| Lard | 65.0 | 59.7 | 5.3 | 75.6 |
| Lard + 2 per cent. of beef fat | 64.4 | 58.8 | 5.6 | 75.6 |
| 5 | 64.0 | 59.3 | 4.7 | 73.4 |
| 10 | 63.9 | 60.3 | 3.6 | 71.1 |
| 20 | 63.3 | 60.5 | 2.8 | 68.9 |
| 40 | 62.6 | 61.1 | 1.5 | 65.6 |
| 60 | 61.8 | 61.2 | 0.6 | 63.0 |
| 80 | 61.4 | 61.0 | 0.4 | 62.2 |
| Beef fat | 61.6 | 60.9 | 0.7 | 63.0 |

The variation in the Bömer values obtained by different workers using the same standardised procedure on the same sample is not unduly large when one considers that differences of only 0.1° C. in each of the two melting-points may result in a difference of 0.5° C. in the value Mg + 2d. With genuine lards or mixtures containing not more than 30 per cent. of adulterant it is unusual for two workers to record values for Mg + 2d differing by as much as 1° C., but with mixtures containing large proportions of adulterant a somewhat greater variation must be admitted.

We have some evidence that with such mixtures some variation results when different quantities of fat are employed. In the method given by the A.O.A.C. there is the direction that "should the volume of crystals materially exceed 3 c.c., take a smaller quantity of lard (3–4 grams) for a new test." From a few experiments we have made it would appear to be equally important to avoid reducing the amount of fat so far that a very small amount of deposit is obtained. This is

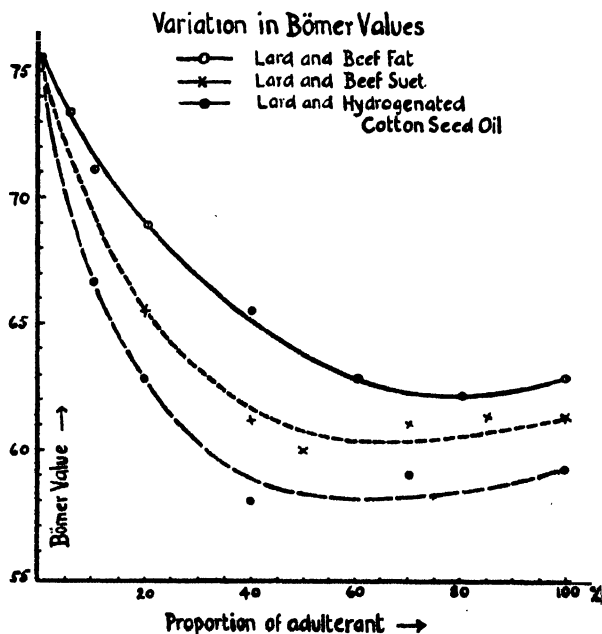
not surprising when one considers the different glycerides which may be deposited from an acetone solution of beef fat. Hilditch and Paul,⁶ in work on the component glycerides of ox depot fat, found that the following glycerides may be deposited from an acetone solution:

| | | |
|---------------------------------------|-----|-------------------------------|
| Most of the tripalmitin .. | 2.8 | from a total of 3.4 per cent. |
| Most of the dipalmito-stearin .. | 5.4 | " " " " 7.8 |
| All the palmito-distearin .. | 5.8 | " " " " 5.8 |
| All the tristearin (if any) .. | 0.4 | " " " " 0.4 |
| Some of the "oleo" dipalmitin .. | 4.9 | " " " " 14.7 |
| Some of the "oleo" palmito-stearin .. | 0.8 | " " " " 32.0 |
| All the "oleo" distearin .. | 2.3 | " " " " 2.3 |
| None of the palmito-di"oleins" .. | 0 | " " " " 23 |
| Some of the stearo-di"oleins" .. | 1.5 | " " " " 10.9 |

Note.—In the above grouping, "palmito" and "palmitin" include the minor component glycerides of myristic, tetradecenoic and hexadecenoic acids.

It seems reasonable to assume that, even with a constant temperature, different concentrations of fat in the solvent will result in a deposition of variable proportions of the different glycerides. With lard, the glyceride structure of which is given by Hilditch and Stainsby,⁷ it would appear that the deposit will consist almost entirely of the palmito-distearin and the dipalmito-stearin.

FIG. 1.



Differences between the melting-points obtained in two laboratories may occur. With certain glycerides and fatty acids a few of the crystals will melt only when the temperature is raised. However, in the method specified by the A.O.A.C. the temperature is recorded when the whole is clear, and we have adopted this procedure throughout. For the determinations we have used the apparatus specified in *Methods of Analysis* (A.O.A.C.), with the addition of a small electric lamp which is shaded and arranged to direct the light through the water in which the tubes are immersed. Duplicate determinations of melting-points in tubes filled from the same batch of material rarely differ by more than 0.1 or 0.2° C.

If a low figure is obtained in the Bömer test a search for iso-oleic acid must be made and the m.p. of the sterol acetates determined. If these tests preclude the presence of hydrogenated oils and fats, the adulterant is beef fat (or mutton or horse fat), and an approximate measure of the proportion of beef fat can be obtained by comparison with known mixtures.

We do not suggest that these curves are exactly reproducible. They will obviously vary to some extent with the particular lard or beef product in the mixtures, and quite clearly only the first portion of the curves may be used. We submit, however, that if the details specified by the A.O.A.C. are closely followed, a Bömer value of 70 indicates the presence of between 5 and 15 per cent. of a beef product, and a value of 65 indicates between 15 and 40 per cent.

THE DETECTION OF HYDROGENATED OILS AND FATS.—Three procedures for determining iso-oleic acid are available, *viz.* separation of the lead salts with ether, the method of Twitchell,²⁶ and the method of Cocks, Christian and Harding.²⁷ For the examination of lard we much prefer the method of Twitchell, since it effects more complete separation of the liquid unsaturated acids. In fact, with genuine lards the iodine values of the "solid" acids separated by Twitchell's procedure are so uniformly low that the test may be used to obtain confirmatory evidence of the presence of substantial amounts of beef fat.

In Twitchell's process two precipitations of the lead salts are made in 95 per cent. alcohol, and this seems to be desirable. It obviously reduces the possibility of obtaining mixed salts containing saturated and unsaturated acids in combination with the same lead atom. In his original paper Twitchell states that with one precipitation the "solid" acids prepared from the insoluble lead salts will have iodine values ranging from 3 to 10, and that no amount of washing of the precipitate will lower this figure. With a second precipitation, however, the iodine value of the "solid" fatty acids is reduced to small proportions. Our results on lard samples are in agreement with this. Hilditch and Priestman²⁸ find that Twitchell's separation results in somewhat low figures for the saturated fatty acids, appreciable quantities of myristic acid (if present), and smaller quantities of palmitic acid, finding their way into the soluble lead salt or "liquid" acid portion. This, of course, is unimportant in the examination of lard for hydrogenated products.

Cocks, Christian and Harding²⁷ state that sometimes they were unable to get consistent results by Twitchell's method, and that the figures for iso-oleic acid were considerably below the truth when the method was applied to certain hydrogenated oils. They recommend an alternative procedure in which there is only one precipitation of the lead salts in 92-93 per cent. alcohol, the insoluble lead salts being filtered off and washed with petroleum spirit. In our hands this method has given unsatisfactory results with lard. On several occasions with known genuine samples the iodine value of the "solid" acids has been as high as 10, and with one lard a value of 14 was obtained. We can only conclude that the "solid" acid fraction is usually contaminated with a substantial amount of "liquid" acids.

For the lead salt and ether method we have followed the details given in Bolton's description²⁹ of the test, with the exception that dilute nitric acid has been used for liberating the fatty acids from the lead salts. Generally speaking, the results have been satisfactory, although we have occasionally encountered explosive boiling (with some loss of the acids) in "refluxing" the lead salts of the "solid" acids with ether. With lard the method has given slightly higher proportions of saturated fatty acids, but the iodine value of the "solid" fraction has tended to be somewhat greater than with Twitchell's method. In a few results with hydrogenated products the percentage of iso-oleic acid obtained after separation by the lead salt and ether method has been slightly less than in the Twitchell method. So far as our experience goes, therefore, we consider that, for our purpose, the greater accuracy in the determination of iso-oleic acid (should it be

present) which may be afforded by other methods must be sacrificed in favour of the more complete removal of the unsaturated "liquid" acids by the Twitchell method. In the examination of lard for the possible presence of small amounts of hydrogenated oil it is our opinion that the method chosen should be the one in which the usual unsaturated acids are most completely removed, so that, if the iodine value of the "solid" acids is appreciable, this shall not indicate an imperfect separation of "liquid" acids, but shall prove the presence of unsaturated solid fatty acids.

Using Twitchell's method on known genuine lards and pig fat, the highest figure we have obtained for the iodine value of the "solid" acids is 2.3, and this was equivalent to an indication of 0.9 per cent. of iso-oleic acid (expressed as a percentage of the total fatty acids). This result was obtained on the specimen of fat rendered from H-bones. More frequently with genuine lards the iodine value was very close to 1.0.

With beef products the iodine value of the "solid" acids is always greater than with lard. Our results have varied from 4.4 to 7.6, and these figures are in agreement with those obtained by other workers.^{26,28} The difference in the figures obtained for genuine lard and beef products appears to be quite significant and, in our opinion, valuable confirmatory evidence can be obtained by using Twitchell's method in the examination of lards which are adulterated with substantial proportions of beef fat.

Table VII contains our results on the specimens of fat rendered from different portions of the pig's carcase (*vide supra*), on a few beef products, and on two samples of "lard" with Bömer values of 63 and 67. The proportions of the acids are expressed as percentages of the total fatty acids.

TABLE VII

| | Solid acids Per Cent. | Iodine value of solid acids | Solid saturated acids Per Cent. | Iso-oleic acid Per Cent. |
|----------------------|-----------------------------|-----------------------------------|--|--------------------------------|
| <i>Pig fat</i> | | | | |
| Back | 40.8 | 0.9 | 40.4 | 0.4 |
| Leaf | 43.0 | 0.7 | 42.7 | 0.3 |
| Midrum | 46.6 | 0.7 | 46.2 | 0.4 |
| Collar | 30.3 | 1.3 | 29.9 | 0.4 |
| Leg bones | 36.2 | 2.0 | 35.4 | 0.8 |
| H-bones | 37.0 | 2.3 | 36.1 | 0.9 |
| <i>Beef products</i> | | | | |
| Beef fat .. | 43.2 | 6.9 | 39.9 | 3.3 |
| " dripping | 47.1 | 4.9 | 44.5 | 2.6 |
| " suet .. | 57.5 | 4.9 | 54.4 | 3.1 |
| Edible beef fat | 55.9 | 7.5 | 51.2 | 4.7 |
| Crude tallow | 50.1 | 7.6 | 45.9 | 4.2 |
| Beef .. No. 7 | 54.4 | 6.0 | 50.8 | 3.6 |
| " " " 8 | 57.4 | 6.9 | 53.0 | 4.4 |
| " marrow fat | 38.8 | 4.4 | 36.9 | 1.9 |
| "Lard" (1) | 43.9 | 5.0 | 41.5 | 2.4 |
| " (2) | 39.0 | 3.1 | 37.7 | 1.3 |

The high results for beef products may presumably be due in part to the presence of some iso-oleic acid, but, probably to a large extent, also to the presence of oleic acid contaminating the "solid" fraction. Grossfeld³⁰ reports the presence of iso-oleic acid in beef fat (1-1.8 per cent.) and in one sample of mutton fat (5.35 per cent.). It is certainly not easy to reduce the iodine value of the solid acids to low dimensions by further solution and reprecipitation. In one of our experiments the solid fatty acids were separated from four samples of the same beef suet. With the first sample two precipitations were made, as in the standard Twitchell method, and with the others, three, four, and five precipitations were made. The iodine values of the "solid" acids so prepared were 5.0, 3.8, 3.0 and 2.6.

In Table VIII the results obtained by Twitchell's method and by the lead salt and ether method are compared, the proportions of acids again being expressed as percentages of the total fatty acids.

TABLE VIII

| | | | | Method | Solid acids Per Cent. | Iodine value of solid acids | Solid saturd. acids Per Cent. | Iso-oleic acid Per Cent. |
|---|--|--|--|---------------------|--------------------------|-----------------------------------|--|--------------------------------|
| Leaf fat | | | | Twitchell | 43.0 | 0.7 | 42.7 | 0.3 |
| | | | | Lead salt and ether | 45.7 | 3.0 | 44.2 | 1.5 |
| | | | | Do. " | 44.4 | 2.0 | 43.4 | 1.0 |
| Pig leg bone | | | | Twitchell | 36.2 | 2.0 | 35.4 | 0.8 |
| | | | | Lead salt and ether | *— | 3.1 | — | — |
| ,, caul fat | | | | Twitchell | 46.6 | 0.7 | 46.2 | 0.4 |
| | | | | Lead salt and ether | 48.2 | 1.4 | 47.4 | 0.8 |
| Beef fat | | | | Twitchell | 43.2 | 6.9 | 39.9 | 3.3 |
| | | | | Lead salt and ether | 47.2 | 9.6 | 42.2 | 5.0 |
| "Lard" (1) | | | | Twitchell | 43.9 | 5.0 | 41.5 | 2.4 |
| | | | | Lead salt and ether | 47.0 | 4.1 | 44.9 | 2.1 |
| ,, (2) | | | | Twitchell | 39.0 | 3.1 | 37.7 | 1.3 |
| | | | | Lead salt and ether | 42.2 | 5.8 | 39.5 | 2.7 |
| Hydrogenated pig fat | | | | Twitchell | 58.6 | 16.6 | 47.8 | 10.8 |
| | | | | Lead salt and ether | 56.6 | 15.7 | 46.7 | 9.9 |
| ,, sesamé oil | | | | Twitchell | 60.4 | 29.5 | 40.6 | 19.8 |
| | | | | Lead salt and ether | 58.8 | 28.2 | 40.4 | 18.4 |
| ,, cotton-seed oil (I.V. 42) | | | | Twitchell | 71.5 | 25.2 | 51.5 | 20.0 |
| | | | | Lead salt and ether | 65.9 | 23.0 | 49.1 | 16.8 |
| ,, cotton-seed oil (I.V. 68) | | | | Twitchell | 42.4 | 34.1 | 26.3 | 16.1 |
| | | | | Lead salt and ether | *— | 33.6 | — | — |

* Some loss of solid acids.

The figures in Table IX indicate that the results obtained in Twitchell's method are reproducible. The acids are expressed as percentages of the total fatty acids. For the first and third samples the duplicate determinations were made by different workers.

TABLE IX

| | | | | Wt. of fatty acids used g. | Solid acids Per Cent. | Iodine value of solid acids | Solid saturated acids Per Cent. | Iso-oleic acid Per Cent. |
|--------------------------------|--|--|--|-------------------------------------|-----------------------------|-----------------------------------|--|--------------------------------|
| Hog fat (commercial sample) .. | | | | 2.019 | 38.0 | 2.2 | 37.1 | 0.9 |
| | | | | 3.276 | 39.1 | 2.5 | 38.0 | 1.1 |
| Pig leg bones | | | | 4.533 | 36.2 | 2.0 | 35.4 | 0.8 |
| | | | | 3.118 | 36.1 | 1.9 | 35.3 | 0.8 |
| Hydrogenated arachis oil .. | | | | 2.996 | 43.5 | 47.2 | 20.7 | 22.8 |
| | | | | 1.939 | 45.3 | 47.4 | 21.4 | 23.9 |
| Beef suet | | | | 2.820 | 56.9 | 5.0 | 53.7 | 3.2 |
| | | | | 3.035 | 57.5 | 4.9 | 54.4 | 3.1 |
| | | | | 3.009 | 56.0 | 5.0 | 52.9 | 3.1 |
| ,, fat | | | | 3.135 | 43.2 | 6.9 | 39.9 | 3.3 |
| | | | | 3.352 | 42.8 | 6.6 | 39.7 | 3.1 |

OTHER TESTS

EXAMINATION OF STEROLS.—A convenient procedure is given by More,³¹ while Hawley³² describes a different technique, in which the digitonide is precipitated in a chloroform solution of the fat. We prefer the former method.

It has been our practice to acetylate the digitonides obtained by More's procedure in a small bottle under pressure, although, according to some workers, boiling with acetic anhydride in an open tube is adequate. It is not necessary to use an oil-bath at 140°C . to remove the excess of acetic anhydride, and in fact such treatment is objectionable. It frequently results in a darkening of the material, and this necessitates treatment with charcoal. Our procedure is to remove the glass stopper from the acetylation bottle, and to substitute a small wash-bottle fitting, one tube of which extends well into the bottle. The other tube, which projects only just below the cork, is connected to a water pump and the bottle is then supported in a small bath of boiling water. The excess acid is soon removed and a very clean acetate is obtained. This is subjected to the usual crystallisation process. We find the Jena micro filter (12 G3—capacity 2 ml.), recommended by Hawley, to be very useful, and with quite small amounts of material it is easily possible to make four or even five recrystallisations.

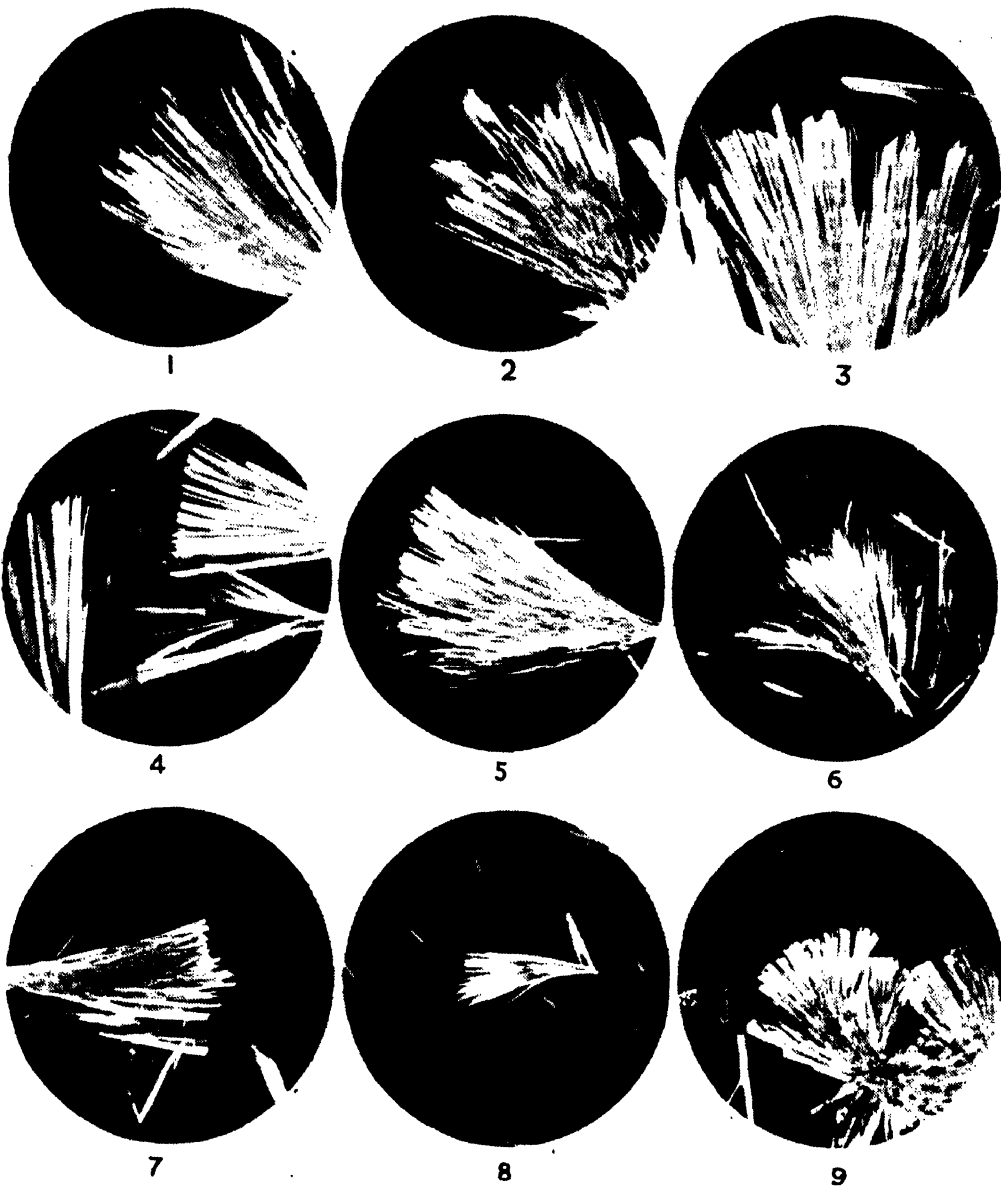
MICROSCOPIC EXAMINATION OF CRYSTALS.—A microscopical examination of the crystals that separate from ether should always be made. The method adopted by some workers of cooling in water or ice water an ethereal solution of the fat leads to poor results. Our procedure is to dissolve 40 drops of the melted fat in 10 ml. of ether in a test-tube, which is then lightly plugged with cotton wool and left for the solution to crystallise at room temperature overnight. Should there be a large deposit it is desirable to begin again with a smaller quantity of fat. Some of the deposit is withdrawn by means of a dipping tube, allowed to fall into a drop of clove oil on a microscope slide, and then covered with a cover-slip. These conditions promote the formation of large crystals, and with genuine lards there is no difficulty in obtaining large well-formed crystals with typical chisel ends.

In the crystals produced under these conditions from known mixtures of beef fat and genuine lard we find a gradual transition from the large chisel-shaped crystals to the typical beef-type crystals and no suggestion of a mixture of both forms. The proportion of beef fat, etc., required to effect the change in crystal form varies with the product used. With beef fat we found that a 40 per cent. admixture was necessary before the chisel ends disappeared completely and typical beef-type crystals were to be observed. With beef suet a 10 per cent. admixture caused some alteration from the typical lard crystals, with 20 per cent. the chisel ends were still just visible, and probably the typical beef crystals would have been obtained with about 30 per cent. admixture. With mixtures made from one sample of beef dripping 10 per cent. caused noticeable alteration from the typical lard crystal, and crystals of the beef type were obtained with a 20 per cent. admixture. These results are illustrated in Plate I.

Certain mixtures of lard with 10 or 20 per cent. of hydrogenated oils gave crystals which could not be distinguished from beef fat, others yielded crystals unlike those from either beef fat or lard. Fortunately such adulterants would usually be detected in the sterol acetate test or in the determination of iso-oleic acid.

Our examination of such mixtures suggests that the genuineness of a sample of lard is not necessarily established when it is possible to observe chisel ends to the crystals. If to find such features a careful search of the slide has to be made and, as is sometimes suggested, the ends of the crystals focussed carefully with the $\frac{1}{4}$ -inch objective, it is almost certain that the lard is adulterated. We have on occasion heard the opinion expressed that if the beef-type crystal is to be found the lard may be adulterated, but that if the crystals have chisel ends the lard is undoubtedly genuine. We incline to the view that when the crystals have chisel ends the lard may be genuine, but that when the beef-type crystal is obtained the lard is undoubtedly adulterated.

PLATE I. CRYSTALS OF LARD AND OF MIXTURES OF LARD WITH BEEF FATS



1. Lard. 2. Lard + 5 per cent. of beef fat. 3. Lard + 10 per cent. of beef fat.
 4. Lard + 20 per cent. of beef fat. 5. Lard + 40 per cent. of beef fat. 6. Beef
 fat. 7. Lard + 20 per cent. of beef suet. 8. Lard + 40 per cent. of beef suet.
 9. Lard + 10 per cent. of beef dripping. Magnification, $\times 70$.

REFERENCES

1. König and Schluckebier, *Z. Unters. Nahr. Genussm.*, 1908, 641.
2. N. R. Ellis and O. G. Hankins, *J. Biol. Chem.*, 1925, 66, 101.
3. N. R. Ellis and H. S. Isbell, *id.*, 1926, 69, 219.
4. ——— *id.*, 1926, 69, 239.
5. N. R. Ellis and J. H. Zeller, *id.*, 1930, 89, 185.
6. R. Bhattacharya and T. P. Hilditch, *Biochem. J.*, 1931, 25, 1954.
7. T. P. Hilditch and W. J. Stainsby, *id.*, 1935, 29, 90.
8. T. P. Hilditch and S. Paul, *id.*, 1938, 32, 1775.
9. W. F. K. Stock, *ANALYST*, 1894, 19, 2.
10. Emery, *U.S. Dept. of Agric.*, 1908.
11. G. D. Elsdon, "*Edible Oils and Fats*," London.
12. A. Bömer, *Z. Unters. Nahr. Genussm.*, 1913, 25, 321; *Abst.*, *ANALYST*, 1913, 38, 204.
13. C. Amberger and A. Wieseahn, *Z. Unters. Nahr. Genussm.*, 1923, 46, 276; *Abst.*, *ANALYST*, 1924, 49, 143.
14. T. P. Hilditch, Private communication.
15. A. Bömer, *Z. Unters. Nahr. Genussm.*, 1913, 26, 559; *Abst.*, *ANALYST*, 1914, 39, 84.
16. H. Sprinkmeyer and A. Diedrichs, *Z. Unters. Nahr. Genussm.*, 1914, 27, 571; *Abst.*, *ANALYST*, 1914, 39, 253.
17. A. Bömer, *Z. Unters. Nahr. Genussm.*, 1922, 43, 87; *Abst.*, *J.S.C.I.*, 1922, 41, 431A.
18. H. Jesser, *Z. angew. Chem.*, 1926, 39, 1551.
19. J. Prescher, *Z. Unters. Nahr. Genussm.*, 1915, 29, 433; *Abst.*, "*Allen's Commercial Organic Analysis*," 5th Ed., Vol. 2, p. 443.
20. E. Polenske, *Abst.*, *ANALYST*, 1907, 32, 382.
21. ——— *Abst.*, *id.*, 1908, 33, 476.
22. Vitoux and C. F. Muttelet, *Ann. Falsif.*, 1920, 13, 593; *Abst.*, *ANALYST*, 1921, 46, 94.
23. ——— *Ann. Chim. anal.*, 1921, 3, 208; *Abst.*, *ANALYST*, 1921, 46, 371.
24. R. H. Kerr, *J.A.O.A.C.*, 1920, 4, 195.
25. *J.A.O.A.C.*, 1937, 20, 276.
26. E. Twitchell, *J. Ind. Eng. Chem.*, 1921, 806.
27. L. V. Cocks, B. C. Christian and G. Harding, *ANALYST*, 1931, 56, 368.
28. T. P. Hilditch and J. Priestman, *id.*, 1931, 56, 354.
29. E. R. Bolton, "*Oils, Fats and Fatty Foods*" (Churchill).
30. J. Grossfeld, *Z. Unters. Lebensm.*, 1931, 62, 553; *Abst.*, *ANALYST*, 1932, 57, 112.
31. A. More, *ANALYST*, 1929, 54, 735.
32. H. Hawley, *id.*, 1933, 58, 530.
33. E. H. Callow, *Report of the Food Investigation Board for the Year 1937*.

DERBYSHIRE COUNTY COUNCIL LABORATORY
COUNTY OFFICES
ST. MARY'S GATE, DERBY

DISCUSSION

The PRESIDENT observed that he had experienced great difficulty in the microscopical examination of the crystals separating from ether, because different forms could be obtained according to the way in which the slide was made and the amount of pressure applied to the cover-glass. He thought that a fair standard would be that the melting-point of the crystals should not be below 63° C.

Mr. K. A. WILLIAMS remarked that the problem before the meeting was one of the most difficult in the analysis of fats. Mr. Sutton had given an admirable account of the methods by which the authors had attacked it, but it was doubtful whether the problem was yet solved. Allegations of adulteration could not generally be refuted by strict evidence of the origin of the fat, and any form of "appeal to the pig" was out of the question. The manufacturer often bought his supplies in the form of crude fat, and whilst he attempted to control his suppliers by carefully-drawn contracts, he could not rely on a warranty, since he refined the material before selling it. For this reason he depended on analysis to check the quality of his raw material, and Mr. Williams quoted one case in which it proved necessary to sample and analyse separately every barrel received in a factory during a period of several months. This placed a heavy burden on the manufacturer, and ruled out the possibility of using any such elaborate investigation as could be made by a local authority on single samples. The manufacturer's difficulties were aggravated by a notable divergence between the views of different experts on the relative merits of various tests. At the date of the meeting it seemed quite sufficient to rely on the B.P. 1932 tests, as presumably these were good enough for the pharmaceutical product and should therefore be satisfactory for a commercial article. If circumstances rendered other testing necessary, Mr. Williams hoped that any method decided upon would be such as could reasonably be applied in factory control. At present there was a "no man's land" in which a pure lard could appear adulterated and an impure lard might be passed. The authors proposed to remove the "no man's

land" by making the standards so stringent that any material falling in it would appear adulterated. This was obviously unfair to the manufacturer, and Mr. Williams trusted that no such principle would be adopted.

He wished to emphasise the point that the manufacturers with whom he had come in contact were concerned to produce a pure product, and worked to what appeared to him to be high standards; but their position was impossible if different authorities required different standards without giving any notice of their preference to the manufacturer.

Mr. Williams showed how the standards suggested in the paper differed radically from those of well-known authorities, although the latter were based on a far wider range of samples than those now proposed. He pointed out how dependent the B.P. crystallisation test was on the exact technique employed, and showed the effect of slight departures from a given form of the test on the melting-point. The latter could be raised by nearly 2 degrees by alteration in the washing technique, and an amplified description was therefore most necessary.

Dealing with the form of the crystals deposited from ether, Mr. Williams demonstrated by lantern slides how both pure and adulterated lards often gave crystals of typically beef-like appearance; but these could usually be resolved into the well-known forms of chisel-shaped or pointed nature by gentle rubbing of the cover-glass on the microscope slide. The false appearance of a beef-like form from pure lard was due to crystallisation in rosettes in which the crystals lay on their edges, and the broad faces of the pure lard crystals could be observed only when the rosettes were broken up. Beef crystals were needle-shaped and softer than lard crystals, so that the rubbing could not make them appear like the latter.

There seemed to be insufficient evidence as yet to prove that a Bömer number of 70 necessarily indicated the presence of 5 to 15 per cent. of beef fat, especially when it was remembered that pigs received a huge variety of diet, the oily constituents of which were often later found in the lard. There were as yet no published data for the effects of many diets on the analytical figures. From the analytical point of view it was also clear that the divergence of 0.2° C. between duplicate melting-points, noted by Mr. Sutton as a reasonable experimental error, led to a possible difference of 1.0° C. in the Bömer number—itsself enough to render the method unsatisfactory.

Mr. Williams therefore preferred, until such time as general agreement could be reached, to use the B.P. standards as being more satisfactory than the authors', and as giving the purchaser all the protection that could reasonably be expected.

Mr. W. M. SEABER mentioned that with certain fats he had been unable to obtain any crystals at all.

Dr. J. H. HAMENCE said that this difficulty in obtaining crystals was a common occurrence with hardened oils. Mixtures consisting entirely of hardened fats crystallised out very slowly—indeed, some would not yield any deposit when crystallisation from ether was attempted under the conditions described for lard in the British Pharmacopoeia. Had Mr. Sutton any experience with the Bömer test when applied to lards with very high iodine values? He (the speaker) had been unable to find anything wrong with some samples which had had iodine values as high as 90, and had reported them to be genuine lards.

Mr. H. E. MONK said that he had not been able to follow how the authors would discover whether an adulterant was hydrogenated fat or beef-fat; the iodine value would not afford much help. Could Mr. Sutton indicate what were the smallest amounts of hydrogenated fat and beef fat that could be detected with reasonable certainty in lard? The photomicrographs shown by the authors were excellent, but he had been astonished to hear that the crystals had been obtained at "room temperature." The slightest variation in technique might produce remarkable differences and rigid control of the temperature and other conditions was necessary if deductions were to be drawn from the form of the crystals.

Dr. C. A. MITCHELL recalled that when Hehner and he were working on flare lard they found that if it were repeatedly recrystallised from ether, beef-like clusters of crystals were obtained, while at the same time the stearic acid content rose with each recrystallisation. He suggested that it might be possible to fix a minimum standard for the proportion of stearic acid in the crystals first obtained.

Mr. R. C. CHIRNSIDE suggested that X-ray patterns of the crystals might afford useful information.

Mr. J. R. NICHOLLS, referring to the question as to which was the best test, said that the Bömer value was essentially an arbitrary accentuation of the result given by the B.P. test and was subject to the equivalent of three experimental errors. He thought that the one value given by the B.P. test was sufficient for deductive purposes. Some years ago a considerable amount of work had been done in the Government Laboratory on the melting-points of the crystals from beef and lard and he had looked up the working sheets. The lowest melting-point for pure lard was 62.7° C. and the highest for pure beef fat was also 62.7° C. obtained after 10 crystallisations. Referring to the crystal form, Mr. Nicholls said that lard crystals should not be mistaken for beef fat crystals; it was well known that, with repeated recrystallisation, one could get beef fat crystals similar to those of lard. Crystals from lard were straight, whilst those from beef fat were usually curved.

Mr. SUTTON, replying, said that in the first place he did not wish to be quoted as suggesting that 57 was the limit for the iodine value. He merely recorded that of the 113 samples examined recently in his laboratory, the iodine value of 96 per cent. of them fell within the range of 57 to 73. Both Professor Hilditch and Mr. Williams had raised the question why the factor $Mg + 2d$

should be chosen. He had not seen any reason for this stated in the literature, but might suggest two points for consideration. First, lard and beef fat differed in both the glyceride melting-point and the "difference" figure, and it therefore seemed proper to take both into consideration in formulating any factor as a criterion of genuineness. Secondly, the figures for d were relatively more divergent, so that it seemed reasonable to use as high a multiple of d as possible. On the other hand, the greater the multiple of d incorporated in the factor the greater was the possible experimental error. $Mg + 2d$ was presumably a compromise giving a factor which indicated most quickly the presence of beef products and at the same time could be duplicated with reasonable agreement.

He could not agree with Mr. Williams about the beef-type crystals which presented a different appearance if rolled over by moving the cover slip. It was not reasonable to assume that all the lard crystals on a slide would be appropriately placed so that only the edges of the crystals would be viewed through the microscope. No manipulation of the cover-slip was necessary or desirable. If the slide were prepared as suggested, one always found the chisel end with genuine lard—in fact, there was no difficulty in recognising a genuine lard crystal. If one did exert pressure on the cover-slip one stood a chance of flattening the crystals. In reply to Dr. Mitchell's statement, he had no experience of the effect of several crystallisations from ether, and his remarks on crystal form referred solely to the standard procedure given in the paper.

He had had several samples with high iodine values and some of these gave insufficient glycerides for the Bömer test. If no glycerides at all were obtained from acetone and adulteration was suspected because of a high iodine value, it could be very certain that beef fat was not present, and he would at once have recourse to the iso-oleic acid test. In deciding on the nature of the adulterant one had to consider all the results. As to the proportion of beef fat that could be detected, he believed that as little as 5 per cent. would usually lower the Bömer figure below the value suggested for genuine lard.

Professor T. P. HILDITCH sent the following written communication:

The Bömer values for differences between melting-points of the glycerides separated from acetone and of their mixed fatty acids depend upon a number of factors which make the interpretation complex and render necessary very close control in the technique employed in carrying out a determination. In the first place, each of the individual mixed triglycerides separated by crystallisation can exist in four polymorphic forms (one vitreous and three— α , β' , β —crystalline), so that it is of the greatest importance to use conditions of crystallisation that will lead to the production of the most stable (β), or at all events the β' , form. Complete melting-point data for simple saturated triglycerides, and for symmetrical and unsymmetrical mixed triglycerides containing two saturated fatty acids, have been given by Malkin with Clarkson, Meara and Carter (*J. Chem. Soc.*, 1934, 666; 1939, 103, 577, 1141, 1512), and these authors also define the necessary crystallisation conditions for production of the stable forms.

Secondly, the presence of minor component acids in the crystallised aggregates from acetone (*e.g.* myristic, oleic, isooleic acids) is probably the determining factor in the glyceride and fatty acid melting-points obtained from different fats. This is appreciated by the authors, who have illustrated the point by details from the only study of an animal (ox) depôt fat* which has yet been published (similar data for two pig depôt fats have now been published; *Biochem. J.*, 1940, 34, 971; *ANALYST*, 1940, 569). In a genuine pig fat there is only about 1 per cent. of combined myristic acid and, owing to the relatively high proportion of oleic acid in the fat, less oleo-disaturated glycerides are present to separate in the fraction least soluble in acetone than in ox depôt and many hydrogenated vegetable fats. Hence the separated glycerides consist, to a greater extent than in other cases, of palmitostearins; the twofold result is a high melting-point for these glycerides and a melting-point for their mixed fatty acids controlled by the proportions of palmitic and stearic (oleic, myristic) acids present. Beef fats contain about 4 per cent. of combined myristic acid and much higher proportions of oleo-disaturated glycerides than pig fat; the result is a lower melting-point of the glyceride conglomerate separated from acetone, although the melting-point of the mixed fatty acids therefrom is much the same as, or may be even slightly higher than, in the corresponding material of a pig fat. This is because the presence of more oleic and myristic acid is compensated for by the higher proportions of stearic to palmitic acid in these beef mixed acid fractions, as compared with those from a pig fat.

Similar complications underly the data for the various hydrogenated fats shown in Table V. In all these, according to the degree of hydrogenation in any one fat, a certain amount of sparingly soluble mono-iso-oleodisaturated glycerides may be expected to be separated in the "Bömer fraction," the melting-point of which it will tend to lower. Groundnut oil, which contains saturated C_{20} , C_{22} and C_{24} acids, and sesame (also, *e.g.* linseed or soya-bean) oil, all of which contain very little palmitic acid, produce fractions least soluble in acetone which will include a high proportion of tristearin (m.p. $71^\circ C.$), formed by hydrogenation of triunsaturated C_{18} glycerides. On the other hand, palm and cottonseed oils, rich in palmitic acid, contain large proportions of palmito-glycerides, and whale oil contains complex mixed glycerides of acids with 16, 18, 20 and 22 carbon

* Even in this detailed study, the small amount of myristic acid has to be included with palmitic acid in considering the component glycerides.

atoms; in these cases, therefore, the melting-point of the least soluble glyceride fraction of the hydrogenated fats is lower, owing to the substantial absence of tristearin and the presence of a greater number of mixed saturated glycerides.

The significance of the expression " $Mg + 2d$ " for the Bömer value appears uncertain to the writer. Clearly it is an attempt to combine in one fraction both the high melting-point of the pig fat glyceride fraction and the difference between the melting-points of the glycerides and the fatty acids. But why $2d$? Why not simply d or, if a multiple be desired, why is double the difference chosen? Since the combination of both the glyceride melting-point and the difference between the melting-points is needed to define the genuineness of a pig fat, would not the end be more clearly attained by recording each separately and assessing the fat by consideration of each value? (For instance, in Table VI, the values in columns 2 and 4 illustrate the effect of the addition of beef fat to lard, in the writer's opinion, much more effectively and rationally than the " $Mg + 2d$ " in column 5.)

Quinaldinic Acid as a Reagent for the Separation of Copper and Cadmium

By A. J. LINDSEY, Ph.D., M.Sc., F.I.C., AND R. J. SHENNAN, M.Sc., A.I.C.

Rây and Bose¹ introduced quinaldinic acid as a reagent for the separation of copper from cadmium, but under the conditions specified we failed to obtain satisfactory results,² and we could not effect a separation by more rigorous pH control with sodium acetate as our buffering agent.³ Recently, Majumdar⁴ has criticised our work on the ground that our procedure differed from that of Rây and Bose, whose results he claims to have substantiated when precipitating from sulphuric acid solutions. Majumdar also states that "the separation of copper from cadmium by free quinaldinic acid in presence of acetic acid has always been found to give high results, owing to co-precipitation of cadmium." This was precisely our contention,³ and is in direct contradiction to Rây and Bose, who claim that "the separation may also be carried out in presence of 2 to 3 ml. of glacial acetic acid."

We have shown that, over the pH range 2.5 to 6.86, copper can be completely precipitated from acetic acid—sodium acetate solutions, and have also observed that the pH range can be extended to pH 1.5 if acetic acid is used alone. We have already stated that the separation of copper from cadmium is impossible in acetic acid solution; Majumdar agrees with this, yet considers it curious that we did not attempt such a separation.⁴

In view of Majumdar's insistence that a successful separation can be made in sulphuric acid solution, we have done further work to explain the divergence between our results and those of Rây and his co-workers.

The determinations of copper in pure copper sulphate solutions were made under the conditions set out by Rây and Bose. A 1 per cent. solution of sodium quinaldinate was used as precipitant, and in every instance the solution was made up to a total volume of 180 ml. A typical selection of the results is given in Table I.

TABLE I

| Copper taken mg. | 2N H_2SO_4 added ml. | Reagent solution ml. | Wt. of ppt. mg. | Copper found mg. | Error mg. |
|---------------------|------------------------------|----------------------------|-----------------------|------------------------|--------------|
| 26.0 | 2 | 20 | 171.2 | 25.62 | -0.38 |
| 26.0 | 5 | 20 | 164.9 | 24.67 | -1.33 |
| 26.0 | 10 | 20 | 133.0 | 19.90 | -6.1 |
| 26.0 | 2 | 30 | 174.1 | 26.02 | +0.02 |
| 26.0 | 5 | 30 | 174.2 | 26.04 | +0.04 |
| 26.0 | 5 | 30 | 174.1 | 26.02 | +0.02 |
| 26.0 | 10 | 30 | 159.6 | 23.89 | -2.11 |
| 26.0 | 10 | 40 | 174.1 | 26.02 | +0.02 |
| 26.0 | 10 | 40 | 173.9 | 26.01 | +0.01 |
| 26.0 | 10 | 50 | 174.2 | 26.04 | +0.04 |

These results indicate that for complete precipitation of copper from solutions containing up to 5 ml. of 2 *N* sulphuric acid a twofold excess of reagent is necessary, while for solutions containing 10 ml. of 2 *N* sulphuric acid a threefold excess is essential. Rây and Bose state that a "slight excess" of reagent is necessary, but give no data, while Majumdar, who does not stress the point, used quantities of reagent conforming approximately with the requirements that we find necessary.

Having established precisely the conditions for complete precipitation of copper, we attempted to separate copper from cadmium on the lines suggested by Rây and Bose. The cadmium used was a solution of the pure sulphate, of which the cadmium content had been determined by electrolysis and by precipitation as quinaldinate. The same reagent was used as in the experiments above. In certain instances the cadmium in the filtrate was determined as quinaldinate. Results are given in Table II.

TABLE II

| Metals taken mg. | 2N H ₂ SO ₄ added ml. | Reagent solution ml. | Wt. of ppt. mg. | Metals found mg. | Error mg. |
|----------------------------|---|----------------------------|-----------------------|------------------------|------------------|
| Cu. 26.00 } Cd. 31.22 } | 2 | 30 | { 178.9 — | 26.77 — | + 0.77 — |
| Cu. 26.00 } Cd. 31.22 } | 5 | 30 | { 178.7 — | 26.74 — | + 0.74 — |
| Cu. 26.00 } Cd. 62.44 } | 10 | 40 | { 176.7 — | 26.43 — | + 0.43 — |
| Cu. 26.00 } Cd. 31.22 } | 2 | 30 | { 179.1 121.9 | 26.80 30.01 | + 0.80 - 1.21 |
| Cu. 26.00 } Cd. 93.66 } | 10 | 40 | { 176.9 376.8 | 26.46 93.05 | + 0.46 - 0.55 |
| Cu. 26.00 } Cd. 31.22 } | 5 | 30 | { 176.4 124.0 | 26.39 30.54 | + 0.39 - 0.68 |
| Cu. 26.00 } Cd. 93.66 } | 2 | 30 | { 176.8 377.4 | 26.45 92.87 | + 0.45 - 0.79 |
| Cu. 26.00 } Cd. 93.66 } | 10 | 40 | { 176.5 377.2 | 26.40 92.87 | + 0.40 - 0.79 |
| Cu. 26.00 } Cd. 62.44 } | 5 | 30 | { 176.5 250.1 | 26.40 61.59 | + 0.40 - 0.85 |
| Cu. 26.00 } Cd. 62.44 } | 10 | 40 | { 177.6 249.7 | 26.58 61.55 | + 0.58 - 0.89 |
| Cu. 52.00 } Cd. 31.22 } | 2 | 60 | { 352.2 — | 53.34 — | + 1.34 — |
| Cu. 26.00 } Cd. 93.66 } | 10 | 40 | { 176.8 — | 26.45 — | + 0.45 — |

In every instance the result for copper was high and that for cadmium correspondingly low. Care was taken in the washing of the copper precipitate to avoid retention of soluble cadmium salts. Tests on the copper quinaldinate precipitates, with "Cadion" as the reagent, showed traces of cadmium. These results completely endorse our previous statement and do not agree with the findings either of Rây and Bose or of Majumdar.

Majumdar (*loc. cit.*) states that copper quinaldinate is appreciably soluble in acetic acid and that copper is completely precipitated from sulphuric acid at pH 1.22; from this he concludes that the solubility of copper quinaldinate in acetic acid is a specific effect and has nothing to do with pH. This conclusion is erroneous. Table I shows that copper quinaldinate is somewhat soluble in sulphuric acid of pH 1.22 (10 ml. of 2 *N* sulphuric acid in 180 ml. solution), but evidently solution can be prevented by employing excess of reagent. In this connection we made the following experiment: An excess of copper quinaldinate

was shaken with sulphuric and acetic acids of various concentrations for a period of 24 hours. The undissolved salt was filtered off, and the copper in solution was determined either electrolytically or colorimetrically.

TABLE III

| 2N H ₂ SO ₄ in 180 ml. of solution ml. | pH | Copper found in solution mg. | Copper quinaldinate dissolved mg. |
|--|-------------|---------------------------------------|--|
| 180 | 0 (approx.) | 101.9 | 680.8 |
| 10 | 1.22 | 3.6 | 24.1 |
| 0.9 | 2.05 | 0.2 | 1.3 |
| Glacial acetic acid in 180 ml. of soln. ml. | | | |
| 45 | 2.05 | 2.0 | 13.4 |
| 35 | 2.2 | 0.4 | 2.7 |
| 21 | 2.5 | 0.3 | 2.0 |

The results given in Table III show that copper quinaldinate is soluble in sulphuric acid solutions and that the solubility is related to the pH.

From Shennan's paper it will be noted that whereas copper is completely precipitated at pH 2.2 when no sodium acetate is present, it is only about half precipitated when this salt is used. It is obvious therefore that Majumdar has misunderstood the reference to the effect of sodium acetate upon the solubility of the precipitate.

REFERENCES

1. P. Rây and M. K. Bose, *Z. anal. Chem.*, 1933, **95**, 400.
2. "Organic Reagents for Metals," 3rd edition, published by Hopkin & Williams, Ltd., p. 104.
3. R. J. Shennan, *ANALYST*, 1939, **64**, 14.
4. A. K. Majumdar, *id.*, 1939, **64**, 874.

THE SIR JOHN CASS TECHNICAL INSTITUTE
JEWRY STREET
LONDON, E.C.3

August, 1940

The Micro-Titrimetric Determination of Carbon Dioxide in Carbonates

By A. F. COLSON, B.Sc., F.I.C.

THE titrimetric method of determining carbon dioxide in carbonates, known as the "shaker" method, has been found to give satisfactory results on the macro-scale, and it has now been adapted to the determination of micro-quantities of carbon dioxide when only a few mg. of material are available.

MACRO METHOD.—The sample is put into a Kjeldahl flask fitted with a rubber stopper carrying (1) a tap funnel, and (2) a glass tube bent at right-angles and connected with a narrow cylindrical vessel containing a mixture of sodium hydroxide and barium chloride solutions. This absorption vessel is provided with a glass tap by means of which the assembled apparatus may be joined to a water pump and evacuated. When this has been done an excess of dilute hydrochloric acid is admitted to the sample, and the reaction vessel is carefully heated until the absorption vessel becomes distinctly hot. During this period the whole apparatus is shaken by hand to facilitate absorption of the liberated carbon dioxide. The absorption vessel is then detached, and the residual alkali is neutralised with

hydrochloric acid after addition of phenolphthalein indicator. Finally, the precipitate of barium carbonate is dissolved in a measured excess of standard hydrochloric acid, and the excess is titrated with standard sodium hydroxide solution, methyl orange being used as indicator.

MICRO METHOD.—Preliminary experiments indicated that the successful adaptation of this method to the requirements of micro-analysis must depend primarily upon the fulfilment of the following conditions:—(1) In contrast to the macro procedure, the contents of the absorption vessel must be protected from atmospheric carbon dioxide until most of the residual alkali has been neutralised. (2) The amount of "dead" space in the apparatus must be reduced to a minimum. (3) Suitable precautions must be taken to ensure that "blank" values given by the reagents, etc., are small in comparison with the amounts of carbon dioxide to be determined.

Of the many types of apparatus constructed the compact form shown in Fig. I most nearly satisfies the specified conditions. The apparatus is made of Pyrex glass and consists essentially of a reaction vessel, A, and an absorption vessel, B, each provided with a suitable tap funnel for the introduction of reagents. It is supported at a convenient height by the three glass rods R_1 , R_2 , R_3 , sealed to the base of the flask B.

Reagents.—The following solutions are required:— N and $N/100$ sodium hydroxide solutions; 10 per cent. barium chloride solution; N , $N/15$ and $N/50$ solutions of hydrochloric acid; methyl red indicator (0.05 per cent. aqueous solution); phenolphthalein indicator (0.05 per cent. solution in 50 per cent. alcohol).

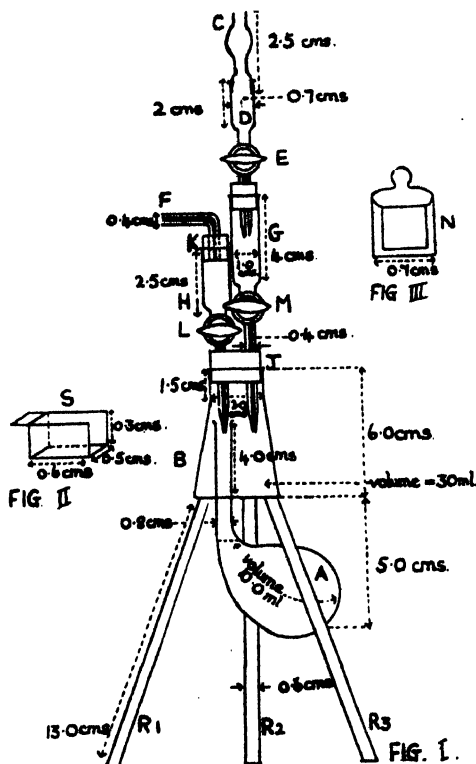
To reduce the "blank" value of the reagents to a minimum the following simple precautions must be taken:—(1) The distilled water used in the preparation of the solutions, and for all other relevant operations, must be boiled in a stream of air free from carbon dioxide and allowed to cool without interruption of the air flow. (2) The N sodium hydroxide solution must be treated with about 0.5 per cent. of barium chloride crystals and allowed to stand for about one day before use.

The strength of the $N/100$ sodium hydroxide solution is best expressed in terms of mg. of the N hydrochloric acid equivalent to 1.0 ml. of the alkali. The N hydrochloric acid solution is standardised against pure sodium carbonate solution and its strength expressed in terms of mg. of carbon dioxide equivalent to 1.0 mg. of the acid.

Procedure.—In making a determination of carbon dioxide the following operations must be carried out in the order given:

(1) The small guard tube C, containing "Carbosorb," is removed from the funnel D, and a little glass wool is introduced into the funnel and pressed down firmly against the tap E. (2) The rubber stopper, which carries the two small funnels G and H, is detached from the neck of the flask B, and the weighed sample, together with its platinum container S (Fig. II) is carefully introduced into the reaction vessel A, and followed by 1.0 ml. of distilled water. (3) The stopper is replaced in such a manner that the tip of the funnel H lies just above the mouth of the reaction vessel. The apparatus must then be rendered air-tight by running a little melted paraffin wax round the rim of the flask B where it meets the rubber stopper. The junctions between the funnel stems and the stopper are similarly sealed. (4) The tube F is now joined to a glass tap through which connection may be made to a water suction-pump, or to a source of air free from carbon dioxide, as required. (5) With the tap E closed, the apparatus is evacuated, filled with air free from carbon dioxide, and again evacuated. (6) 0.25 ml. of 10 per cent. barium chloride solution is introduced into the funnel D and drawn through the glass wool filter pad into the funnel G. It is followed by 0.25 ml. of N sodium hydroxide solution and about four drops of phenolphthalein indicator solution. The perfectly clear solution is then allowed to enter the flask B, and the funnels D and G are rinsed through with about 0.5 ml. of water, applied in several small

portions. The funnel G is finally rinsed out with *N* hydrochloric acid and water, the rinsings being rejected. (7) 0.3 ml. of *N* hydrochloric acid and one or two drops of methyl red indicator solution are admitted from the funnel H to the sample in the reaction vessel A. This volume of acid is sufficient for any 5 mg. sample.



(8) The reaction vessel, A, is now carefully heated over the flame of a micro burner until the solution boils, and heating is continued until the temperature of the absorption vessel, B, has risen to about 70° C. The apparatus is then allowed to cool for a few seconds, and alternate heating and cooling are continued until only one or two drops of liquid remain in the reaction vessel. During this process the contents of the vessels A and B must be almost continuously agitated to facilitate absorption of the liberated carbon dioxide and to prevent loss of solution from the reaction vessel by spitting. Agitation of the solutions is conveniently effected by holding the supports R_1 , R_2 , R_3 in the right hand, and imparting a gentle circular movement to the apparatus, while pressing the three supports firmly against a smooth bench surface.

(9) The reaction vessel is next filled with distilled water from the funnel H, to a point about two-thirds up the narrow neck. (10) A stream of air free from carbon dioxide is passed through the tube F attached to the rubber stopper K, and, without interruption of the air flow, the stopper K is inserted into the funnel H, which has previously been completely filled with water. By opening the tap L the apparatus is filled with the purified air. The tap L is then closed and the apparatus is shaken gently for two or three minutes. (11) The apparatus is next partially evacuated, and the residual alkali in the absorption vessel is nearly neutralised by dropwise addition of *N*/15 hydrochloric acid, from the funnel G.

(12) The tap M is closed, and the apparatus is filled with air free from carbon dioxide by opening the tap L after connecting the tube F with the supply of air. (13) The rubber stopper is then removed, and the wall of the absorption vessel is rinsed down three or four times with small quantities of water from a small wash-bottle with a fine jet. The solution is then exactly neutralised with *N*/50 hydrochloric acid. (14) A weighed excess of *N* hydrochloric acid (about 160 mg.) contained in the small stoppered glass vessel N (Fig. III) is introduced into the vessel B, and followed by two drops of methyl red indicator solution, and the solution is boiled gently for five minutes over the flame of a micro burner. (15) Finally, the flask is closed with a rubber stopper and cooled by standing the apparatus in a large beaker of cold water, and the excess of hydrochloric acid is titrated with *N*/100 sodium hydroxide solution. From the volume of alkali required, the weight of *N* acid used to react with the barium carbonate is obtained, and the carbon dioxide content of the sample is calculated. The time taken to carry out a complete determination is about one hour. Before the actual determinations a "blank" test must be made under exactly the same conditions. It is convenient for the calculation to express the value of the "blank" in terms of mg. of *N* hydrochloric acid.

Results.—Selections from a large number of results obtained in the determination of approximately 1.0, 0.5 and 0.25 mg. of carbon dioxide in specially prepared pure barium carbonate are given in Table I. By the macro "shaker" method this pure barium carbonate was found to contain 99.72 per cent. of carbon dioxide, and this value was used throughout in calculating the weights of carbon dioxide recorded in the third column of the table.

TABLE I
DETERMINATION OF CARBON DIOXIDE IN BARIUM CARBONATE

| Expt. No. | BaCO ₃ taken mg. | CO ₂ present mg. | CO ₂ found mg. | Error mg. CO ₂ | Error Per Cent. |
|-----------|--------------------------------|--------------------------------|------------------------------|------------------------------|--------------------|
| A1 | 4.814 | 1.070 | 1.057 | -0.013 | -1.20 |
| A2 | 5.058 | 1.124 | 1.136 | +0.012 | +1.10 |
| A3 | 4.825 | 1.073 | 1.045 | -0.028 | -2.60 |
| A4 | 4.782 | 1.063 | 1.107 | +0.044 | +4.10 |
| A5 | 5.063 | 1.126 | 1.124 | -0.002 | -0.20 |
| B1 | 2.607 | 0.580 | 0.575 | -0.005 | -0.90 |
| B2 | 2.470 | 0.549 | 0.553 | +0.004 | +0.70 |
| B3 | 2.588 | 0.575 | 0.554 | -0.021 | -3.70 |
| B4 | 2.444 | 0.543 | 0.520 | -0.023 | -4.20 |
| C1 | 1.250 | 0.278 | 0.266 | -0.012 | -4.30 |
| C2 | 1.293 | 0.288 | 0.278 | -0.010 | -3.50 |
| C3 | 1.190 | 0.265 | 0.250 | -0.015 | -5.70 |
| C4 | 1.220 | 0.271 | 0.257 | -0.014 | -5.20 |

DISCUSSION OF SOURCES OF ERROR.—Some of the operations described above were designed to eliminate or reduce the errors derived from various sources. The filtration of the *N* sodium hydroxide solution through the glass wool pad contained in the funnel D, serves to remove any suspended barium carbonate that may be present in the reagent. Any traces of barium carbonate deposited in the funnel G would subsequently be dissolved by the *N*/15 hydrochloric acid used for the neutralisation of the residual alkali in the absorption vessel, and in consequence this acid would contain a little dissolved carbon dioxide, which would give rise to a positive error if it were allowed to enter the alkaline solution in the absorption vessel. The funnel G must, therefore, be rinsed out with dilute acid and water before the introduction of the *N*/15 hydrochloric acid.

The importance of excluding air from the interior of the apparatus until nearly

the whole of the residual alkali has been neutralised is clearly illustrated by the following "blank" determinations. In Expt. No. 1 the neutralisation of the alkali was carried out after the absorption flask B had been opened by removal of the stopper. The two values obtained were high and in poor agreement. In Expt. No. 2 the neutralisation was effected without opening the absorption vessel. The values thus obtained were lower and much more concordant. In Expt. No. 3 the apparatus was kept closed and also sealed with paraffin wax as described above. The two "blank" values obtained were lower than those in Expts. Nos. 1 and 2, and were in close agreement. (See Table II.)

TABLE II

"Blank" value

| Expt. No. | N HCl mg. | CO ₂ mg. |
|-----------|--------------|------------------------|
| 1 | 11.90 | 0.262 |
| | 14.80 | 0.326 |
| 2 | 3.48 | 0.077 |
| | 3.36 | 0.074 |
| 3 | 1.60 | 0.035 |
| | 1.40 | 0.031 |

For several reasons it would be preferable to use a ground-glass joint in place of the rubber stopper J, but in practice it was found that such joints possess the serious disadvantage of frequently "seizing up." Diffusion of carbon dioxide into the rubber stopper J is a possible source of error, to prevent which the rubber stopper is impregnated with a mixture of paraffin wax and vaseline in the following manner: The stopper is immersed in a melted mixture of paraffin wax and vaseline, contained in a thick-walled round-bottomed flask attached to a good water pump. The flask is evacuated and then air is admitted. Alternate evacuation and admission of air are repeated until air bubbles no longer rise from the surface of the rubber. Finally, the stopper is removed from the flask and the excess of wax is carefully wiped off (*cf.* Pregl, *Quantitative Organic Microanalysis*, 3rd English Ed., p. 26). In the course of a determination this stopper is subject to the action of the steam which enters the absorption flask B, and a little paraffin wax is carried down into the sodium hydroxide solution. Experiments to determine the effect of this impurity indicate that it does not increase the "blank" values by more than 0.01 mg. of carbon dioxide. The solubility of carbon dioxide in the acid solution contained in the reaction vessel A, may give rise to a small negative error, and that solution is therefore distilled almost to dryness to expel as much as possible of the dissolved gas.

It is desirable to reduce the amount of "dead" space in the apparatus as much as possible. For this reason the reaction vessel is partly filled with water at a suitable stage in the determination. After the residual alkali in the absorption flask has been almost completely neutralised by *N*/15 hydrochloric acid, the flask is opened and rinsed down with water. Any hydrochloric acid adhering to the sides of the flask is therefore carried down into the slightly alkaline solution. Experiments indicated that the amount of hydrochloric acid thus introduced is not likely to exceed 0.1 ml. of *N*/100 hydrochloric acid. Since the neutralisation of the residual alkali is always interrupted when at least 0.05 ml. of *N*/15 hydrochloric acid is still required, this additional 0.1 ml. of *N*/100 acid can have no harmful effect. The shape of the vessel employed to contain the weighed sample appears to be of some importance. A small circular glass dish, 4 mm. deep and 4 mm. in internal diameter, was formerly used, but more accurate results are obtained with the platinum scoop S, illustrated in Fig. II. Apparently, when the small glass dish is employed, the whole of the sample does not always come into contact with the acid in the reaction vessel.

The concentrations of the acid and alkali solutions used in this method were chosen with the object of reducing the volume of the apparatus without undue sacrifice of accuracy. For this reason, also, weighed amounts of *N* hydrochloric acid solution are used in preference to larger measured volumes of more dilute acid. The error involved in weighing out the required amount of standard acid does not exceed 0.1 mg., which corresponds to approximately 0.002 mg. of carbon dioxide. The exact neutralisation of the residual alkali in the absorption vessel is facilitated by the following simple device employed by A. Benedetti-Pichler (*Z. anal. Chem.*, 1928, p. 200) and others. By applying a thin film of paraffin wax to the tip of the micro-burette, very small drops of solution can be collected at the orifice, and transferred to the titration vessel by allowing them to touch the wall of that vessel. Additions of 0.01 ml. to 0.03 ml. of 0.02 *N* hydrochloric acid can thus be made. The same technique is employed in the final titration with *N*/100 sodium hydroxide solution. In this titration it is desirable to work with the same portion of the burette for the actual determination of carbon dioxide and for the "blank." It is also advisable to allow a constant drainage time of about two minutes before the final burette readings are taken.

SUMMARY.—A method is described for the micro-titrimetric determination of carbon dioxide in carbonates. Various sources of error are discussed in detail, and a number of results are given which indicate that quantities of 1.0, 0.5 and 0.25 mg. of carbon dioxide may be determined with a deviation of not more than about 1.5, 2 and 5 per cent. respectively from the correct values.

I wish to express my thanks to the Directors of Imperial Chemical Industries, Ltd., for permission to publish this work, which was carried out in the Research Laboratory of their subsidiary Company, Imperial Chemical Industries (Alkali), Ltd., Northwich.

RESEARCH DEPARTMENT

I.C.I., WINNINGTON

NORTHWICH, CHESHIRE

December, 1939

Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

THE CYANOMETRIC DETERMINATION OF COBALT AND NICKEL*

A CYANOMETRIC process for determining cobalt and nickel in presence of each other has been published by B. S. Evans (*ANALYST*, 1937, 62, 363). This method, although excellent, has one or two disadvantages: first, ammonia cannot be used in the initial stage of the titration; secondly, there is a fairly large interval between the first and second stages, which necessitates the burettes being left during that time.

It has been found that when the cyanide is added to the cobalt solution, previously acidified with dilute nitric acid, and the liquid is allowed to stand for at least four minutes with occasional shaking, the cobalt proceeds to the Co:5KCN stage, whereas nickel remains at the Ni:4KCN stage. The following conditions have been worked out for determining cobalt, or cobalt and nickel together: The test sample should contain not more than 0.04 g. of cobalt and nickel, and the volume of liquid should be 150 ml. and approximately neutral. The solution is treated with 10 ml. of nitric acid (sp.gr. 1.2), and with potassium cyanide in excess and allowed to stand for four minutes with occasional shaking. Twenty ml. of dilute ammonia (1/1) (sufficient to neutralise the nitric acid and to leave an excess of about 10 ml.) and 10 ml. of potassium iodide solution (4 per cent.) are added, and the titration is carried out in the ordinary way.

The following results were obtained for cobalt alone:

| Cobalt taken g. | Titration ml. | Cobalt found g. |
|--------------------|----------------------|--------------------|
| 0.0200 | 35.00 — 9.90 = 25.10 | 0.0202 |
| 0.0200 | 34.80 — 9.60 = 25.20 | 0.0202 |
| 0.0200 | 34.80 — 9.10 = 25.20 | 0.0202 |

* Communication from the Research Department, Royal Arsenal, Woolwich.

The following results were obtained for cobalt and nickel together:

| Cobalt taken g. | Nickel taken g. | Total titration ml. | Titration for nickel ml. | Titre for cobalt ml. | Nickel found g. | Cobalt found g. |
|-----------------|-----------------|---------------------|--------------------------|----------------------|-----------------|-----------------|
| 0.0100 | 0.0100 | 34.90—12.10=22.80 | 15.80—5.70=10.10 | 22.80—10.10 | 0.0101 | 0.0102 |
| 0.0100 | 0.0100 | 35.00—12.50=22.50 | 16.30—6.20=10.10 | 22.50—10.10 | 0.0101 | 0.0100 |

In these tests the total cyanide used was measured, and then to an exactly similar quantity of cobalt and nickel, acidified with nitric acid, an excess of cyanide was added to bring cobalt to the Co : 5KCN stage. Excess of cyanide was then destroyed by boiling with 15 ml. of ammonia (1/1) and 25 ml. of hydrogen peroxide (20 vols.) for about 20 minutes. Under these conditions the cobalt proceeds to the Co : 6KCN stage, forming a stable compound. Nickel is then precipitated, free from cobalt, with solid dimethyl-glyoxime, and determined as usual. In all the determinations the factors were 0.001 for nickel, and 0.000803 for cobalt.

The concentration of the nitric acid in the initial stage of transforming cobalt to the Co : 5KCN condition has some importance. For example, with volume 250 ml. instead of 150 ml. the following results were obtained:

| Cobalt taken g. | Cobalt found g. |
|-----------------|-----------------|
| 0.0200 | 0.0197 |
| 0.0200 | 0.0196 |
| 0.0200 | 0.0198 |

The inference is that the concentration of the nitric acid was too low for transformation of the whole of the cobalt to the Co : 5KCN stage in the time stated.

W. J. AGNEW
October, 1940

THE USE OF ANHYDRITE AS A DESICCATING AGENT

Two papers by Hammond and Withrow (*Ind. Eng. Chem.*, 1930, 653 and 1112) have not received, in England, the attention that they deserve; they were not abstracted in *THE ANALYST*. These authors claim that anhydrite is the cheapest and most satisfactory chemical for laboratory use as a desiccating agent. It has been in regular use in this laboratory for over a year, and these claims have been found to be fully warranted.

To prepare anhydrite, commercial gypsum—selected white lumps—is broken up and sieved. The portion that passes an I.M.M. size-5 sieve and is retained by size-10 is used. This is spread out to a depth of about 1 inch in shallow metal trays, which are heated to 230–250° C. (Hammond and Withrow say that 250° C. must not be exceeded) in an electric oven with automatic temperature control. The heating is continued for one hour after a test of the oven vent with a cold watchglass shows that no more moisture is being evolved, and the product is then transferred while hot to tins with well-fitting lids and subsequently bottled. Fresh gypsum requires about 4 hours' heating; it does not undergo any visible change in the process. The anhydrite will absorb about 6 per cent. of its weight of water, and when exhausted can be reactivated by again heating to 230–250° C.

By weighing at intervals a small quantity exposed to the air in a flat dish, it has been found that the absorption of moisture is rapid and even; there is no tailing off in the rate of absorption until over 5 per cent. of the weight of the anhydrite has been absorbed. Anhydrite is a more powerful dehydrating agent than fused calcium chloride. Ten g. of calcium chloride were exposed to the air until it had taken up water equal to about 5 per cent. of its weight. It was then put in a vacuum desiccator over anhydrite for 24 hours; more than half the absorbed moisture was removed. Hammond and Withrow claim that anhydrite will also remove water from sulphuric acid, but this could not be confirmed. Possibly, the vapour pressure of anhydrite is higher than that of moist sulphuric acid at tropical temperatures but lower in more temperate climates.

Anhydrite is very clean in use. Desiccators and gas drying tubes can be emptied and refilled in a moment without the necessity for washing out and drying that the use of calcium chloride or sulphuric acid involves. As with sulphuric acid, it is not possible to tell by appearance when the absorbing power is falling off, but the preparation of a new supply or the reactivation of the old is so cheap and simple that there is little temptation to keep it in use when it may be approaching exhaustion. If necessary, the activity is easily tested by placing a little in a test-tube surrounding a thermometer and adding a few drops of water. If the material is still active, there is an immediate rise in temperature.

Anhydrite is an ideal agent for removing the last trace of water from organic liquids and can be used for drying alcohol. Three hundred ml. of commercial absolute alcohol (about 98 per cent.) were added to 300 g. of anhydrite. There was an immediate rise in temperature. The alcohol was distilled off, and the distillate was found to contain 99.8 per cent. of alcohol.

The authors state that anhydrite owes its rapid action to the fact that it is denser than gypsum. There is no shrinkage during the change to anhydrite, and accordingly the grains are porous.

GOVERNMENT ANALYST'S LABORATORY
MADRAS

HERBERT HAWLEY

October, 1940

ALUMINA FOR THE ESTIMATION OF CAROTENE

IN my paper on Carotene and Allied Substances, published in THE ANALYST (1940, 65, 266), when describing my "3 per cent. acetone" chromatographic process, I mentioned that I had had difficulty in obtaining an alumina that would give results similar to those obtained with Merck's alumina.

Since then I have tried one recently put on the market by Messrs. Savory & Moore. This appears to act very similarly to Merck's. The following are typical comparative results:

| | <i>Dried Grasses</i> | | |
|-----|---------------------------------------|-------------------------------|---|
| | "Crude carotene" parts per million | Carotene (Merck's alumina) | Carotene (Savory & Moore's alumina) |
| (1) | 250 | 155 | 165 |
| (2) | 230 | 190 | 185 |
| (3) | 410 | 360 | 360 |
| | <i>Dried Tomatoes</i> | | |
| | 250 | 40 | 40 |
| | (most of this is lycopene) | | |
| | <i>Dried spent Tea Leaves</i> | | |
| | 105 | 62 | 66 |

It should be pointed out that the alumina readily takes up moisture, and it is desirable to heat it over a very low flame shortly before the tests are made.

W. M. SEABER

ANALYTICAL LABORATORY
OAK AVENUE

HAMPTON, MIDDLESEX

October 28th, 1940

Erratum. November issue, p. 596 (Detection of Beef Fat in Lard), last line but two of the Note. For " $\pm 0^{\circ}\text{C.}$ " read " $\pm 0.1^{\circ}\text{C.}$ "

Ministry of Food

Emergency Powers (Defence)

STATUTORY RULES AND ORDERS. 1940. No. 1748

Food (Compound and Mixed Feeding Stuffs)

Order, dated September 27, 1940, amending the Compound and Mixed Feeding Stuffs (Control) (No. 2) Order, 1940*

THIS Order (which came into force on October 7th) amends "the Principal Order"^a as follows:

- (a) by inserting in Article 1 thereof immediately after the definition of "Licensed concentrate" the following definition:

"Licensed molassed feeding stuff" means a molassed feeding stuff which a person is authorised to manufacture for sale or prepare for sale in accordance with the terms of a licence granted under this Order."

- (b) by inserting in such Article immediately after the definition of "Low protein oilseed cake or meal" the following definition:

"Molassed feeding stuff" means a feeding stuff (other than a compound or concentrate as they are defined in the Feeding Stuffs (Maximum Prices) Order, 1940,^b or a concentrate, as therein defined) which contains not less than 20 per cent. of molasses."

* H.M. Stationery Office, York House, Kingsway, London, W.C.2. Price 4d. net.

^a S.R. & O., 1940, No. 1119 (*cf.* ANALYST, 1940, 505). ^b S.R. & O., 1940, No. 11.

- (c) by substituting for paragraph (c) of Article 2 of that Order the following paragraph:
 "(c) Use in the manufacture or preparation of any compound or mixed feeding stuff described in the first column of Part A of the First Schedule hereto any ingredient other than an ingredient specified in Part A of the Second Schedule hereto or use as aforesaid any ingredient mentioned in the first column of Part B of the said Second Schedule except as specified opposite to such ingredient in the second column thereof."
- (d) by substituting for the First and Second Schedules to that Order the First and Second Schedules to this Order.

THE FIRST SCHEDULE

PART A.—COMPOUND CAKES AND MEALS

The Compound or Mixed Feeding Stuff must comply with the following conditions as to percentage contents (Third Column of Table).

CATTLE AND SHEEP FOODS.

National Cattle Food, No. 1 (Dairy ration or cattle or sheep fattening ration).—Oil, min. 4, max. 6; albuminoid, min. 19.5, max. 21; fibre, max. 9.5.

National Cattle Food, No. 2 (Ration for grass feeding or fattening).—Oil, min. 4, max. 6; albuminoid, min. 14, max. 17; fibre, max. 12.

National Cattle Food, No. 3 (*Rearing*) (Ration for rearing young stock).—Oil, min. 4.5, max. 6; albuminoid, min. 20, max. 22; fibre, max. 8.

National Calf Gruel.—Oil, min. 6, max. 12; albuminoid, min. 15, max. 24; fibre, max. 7.

Each of the four preceding compounds shall contain not more than 2.5 per cent. of lime (calc. as CaCO_3) exclusive of lime in the form of bone flour; not more than 1.5 per cent. of lime in the form of bone flour, and not more than 1.5 per cent. of salt (calc. as NaCl).

PIG FOODS.

National Pig Food, No. 1 (Pig nuts or pig meal for sows or weaners).—Oil, min. —, max. 4; albuminoid, min. 16, max. 18; fibre, max. 7.

The compound shall contain the following percentages of ingredients (Fourth Column of Table):—Maize meal and/or barley meal and/or dried potato products, min. 35, max. —; wheat feed (other than bran), min. 20, max. —; oats, min. —, max. 10; bran, min. —, max. 10; low protein oilseed cake and/or meal, min. —, max. 20; *fish and/or animal protein rich substances, min. 5, max. —; molasses, min. —, max. 5; *sundries including vitamin potent substances and mineral matter at maker's discretion provided that the salt-content of the compound shall not exceed 0.5 per cent., min. —, max. 10; together with sufficient *vegetable protein to ensure that the albuminoid-content agrees with the requirements given above.

National Pig Food, No. 2 (Pig nuts or pig meal for fattening).—Oil, min. —, max. 4; albuminoid, min. 13, max. 15; fibre, max. 7. Percentage ingredients as follows:—Maize meal and/or barley meal and/or dried potato products, min. 35, max. —; wheat feed, min. 25, max. —; ground oats and/or bran, min. —, max. 20; low protein oilseed cakes and/or meals, min. —, max. 25; *fish and/or animal protein rich substances, min. 2.5, max. —; molasses, min. —, max. 5; *sundries (as for No. 1, *supra*), plus *vegetable protein.

POULTRY FOODS (Mash or Pellets).

National Poultry Food, No. 1 (*Summer Laying*) (Laying mash or pellets).—Oil, min. 3, max. —; albuminoid, min. 17, max. 19; fibre, max. 8. Percentage composition:—Wheat feed and/or low protein oilseed cake and/or meal, min. 40, max. 65; cereals, min. 25, max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *sundries including vitamin potent substances and mineral matter at maker's discretion, provided that the salt-content of the compound (calc. as NaCl) does not exceed 1 per cent., min. —, max. 7.5; plus *vegetable proteins to ensure specified albuminoid-content. No decorticated or undecorticated cotton-seed cake or meal may be used.

National Poultry Food, No. 1A (*Winter Laying*).—Oil, min. 3; max. —; albuminoid, min. 17, max. 19; fibre, max. 8. Percentage composition:—Wheat feed and/or low protein oilseed cake and/or meal, min. 40, max. 65; cereals, min. 25, max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *cod-liver oil equiv. to not less than 1 per cent. (on the weight of the compound) of cod-liver oil containing 85 international units of vitamin D per g.; *sundries (as for National Poultry Food, No. 1), min. —, max. 7.5; plus *vegetable protein as necessary for albuminoid-content. No decorticated or undecorticated cotton-seed cake or meal may be used.

National Poultry Food, No. 2 (*Summer Growing*).—Oil, min. 3, max. —; albuminoid, min. 14, max. 17; fibre, max. 8. Percentage composition:—Wheat feed and/or low protein oil seed cake and/or meal, min. 40, max. 65; cereals, min. 25; max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *sundries (as for National Poultry Food, No. 1), min. —, max. 7.5; plus *vegetable protein as necessary for albuminoid-content. No decorticated or undecorticated cotton-seed cake or meal may be used.

* These ingredients may be in the form of licensed concentrates.

National Poultry Food, No. 2A (Winter Growing).—Oil, min. 3, max. —; albuminoid, min. 14 max. 17; fibre, max. 8. Percentage composition:—Wheat feed and/or low protein oil seed cake and/or meal, min. 40, max. 65; cereals, min. 25, max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *cod-liver oil equiv. to 1 per cent. (on weight of compound) of cod-liver oil containing 85 international units of vitamin D per g.; *sundries (as for National Poultry Food, No. 1), min. —, max. 7.5; plus *vegetable protein as necessary for albuminoid-content. No decorticated or undecorticated cotton-seed cake or meal may be used.

National Poultry Food, No. 3 (Battery).—Oil, min. 3, max. —; albuminoid, min. 15, max. 18; fibre, max. 8. Percentage composition:—Wheat meal and/or low protein oilseed cake and/or meal, min. 35, max. 65; cereals, min. 30, max. 50; *fish and/or animal protein rich substance, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *cod-liver oil (as for National Poultry Food, No. 2A, *supra*); *sundries (as for National Poultry Food, No. 1), min. —, max. 15; plus *vegetable protein as necessary for albuminoid-content. No decorticated or undecorticated cotton-seed cake or meal may be used.

National Baby Chick Food.—Oil, min. 3, max. —; albuminoid, min. 16, max. 18; fibre, max. 6.5. Percentage composition:—Wheat feed and/or low protein oilseed cake and/or meal, min. 35, max. 65; cereals, min. 30, max. 50; *fish and/or animal protein rich substances, min. 5, max. 10; molasses (for pellets only), min. —, max. 5; *cod-liver oil (as for National Poultry Food, No. 2A, *supra*); *sundries (as for National Poultry Food, No. 1, except that salt must not exceed 0.75 per cent.), min. —, max. 20; *vegetable protein as necessary for albuminoid-content. Rye or rye products may not be used. No decorticated or undecorticated cotton-seed cake or meal may be used.

NOTE.—(1) The conditions as to oil, albuminoid and fibre contents found on analysis are subject to the limits of variation specified in the Fertilisers and Feeding Stuffs Regulations, 1932.

(2) Where cod-liver oil is prescribed, substances other than cod-liver oil may be used, provided that : (a) the vitamin D content of such substances in the aggregate is not less than that prescribed for cod-liver oil; (b) such substances are warranted in writing by the maker thereof as fully effective for poultry in accordance with the Chick Test of the British Standards Institution.

PART B. CEREAL MIXTURES. PERCENTAGE COMPOSITION (Third Column of Table)

National Cereal Mixture, No. 1.—Barley meal, min. 15, max. 30; maize meal, min. 45, max. 60; fine wheat feed, and/or imported middlings and/or imported pollards, min. 15, max. 25.

National Cereal Mixture, No. 2.—Maize meal, min. 35, max. 55; cattle ground oats, min. 30, max. —; fine wheat feed, and/or imported middlings and/or imported pollards, min. 10, max. 15.

National Poultry Corn, No. 1A.—Wheat and/or barley, min. 15, max. 30; oats, min. 20, max. 45; cut or kibbled maize, min. 40, max. 60.

National Poultry Corn, No. 1B.—Wheat and/or barley, min. 15, max. 30; clipped oats, min. 20, max. —; cut or kibbled maize, min. 40, max. 60.

National Poultry Corn, No. 2A.—Wheat and/or barley, min. 15, max. 30; oats, min. 20, max. —; whole maize, min. 40, max. 60.

National Poultry Corn, No. 2B.—Wheat and/or barley, min. 15, max. 30; clipped oats, min. 20, max. —; whole maize, min. 40, max. 60.

National Chick Feed, No. 1.—Fine cut wheat, min. 20, max. 45; No. 4 maize grits, min. 45, max. 70; cut groats, min. 10, max. —.

National Chick Feed, No. 2.—Cut wheat, min. 20, max. 40; No. 3 maize grits, min. 30, max. 60; whole groats, min. 10, max. —; dari and/or millet, min. —, max. 30.

National Chick Feed, No. 3.—Fine cut wheat, min. 50, max. —; No. 4 maize grits, min. 30, max. —.

National Chick Feed, No. 4.—Cut wheat, min. 50; No. 3 maize grits, min. 25, max. 45; whole groats, min. 10, max. —.

THE SECOND SCHEDULE

PART A

Any feeding stuff contained in the First Schedule to the Feeding Stuffs (Maximum Prices) Order, 1940, as amended, and any of the following feeding stuffs:

Alfalfa meal, biscuit meal, buckwheat (whole, crushed or ground), cod-liver oil or other vitamin potent substances, dari (whole, crushed or ground), distillery dreg meal, dredge corn (whole, crushed or ground), dried butter milk, dried clover meal, dried grass meal, dried liver meal, dried milk, dried yeast, feeding dried blood, groats, herbs, home-grown barley, oats, peas or wheat (whole, crushed or ground), Kaffir corn (whole, crushed or ground), licensed concentrates, licensed molassed feeding stuffs, liquorice-root, lucerne meal, malt, millet (whole, crushed

* These ingredients may be in the form of licensed concentrates.

or ground), mineral matter, rye (whole, crushed or ground), spices, sugar cane molasses, tares (whole, crushed or ground), wheat germ, whey paste, whey powder.

PART B

| First Column Ingredient | Second Column Permitted use |
|--|--|
| 1. Cocoa shell, husk, bran or waste. | Use in cattle or sheep foods (in quantities not exceeding 2.5 per cent. of the completed compound or mixed feeding stuff). |
| 2. Dried milk, dried butter milk or whey powder. | Use in calf gruels and baby chick foods. |
| 3. Wheat, whole, crushed or ground. | Use in poultry and chick foods. |

STATUTORY RULES AND ORDERS. 1940. No. 1898

Raw Materials (Fertilisers)*

THIS Order revokes the Control of Fertilisers (No. 1) Order, 1939, Direction No. 5,^a and fixes the basic prices of superphosphate of lime, ground phosphate and compound fertilisers, for the months of November and December, 1940, and the first six months of 1941.

STATUTORY RULES AND ORDERS. 1940. No. 1899

Raw Materials (Fertilisers)†

THE CONTROL OF FERTILISERS (No. 7) ORDER, 1940, DATED OCTOBER 28, 1940, MADE BY THE MINISTER OF SUPPLY UNDER REGULATIONS 55 AND 98 OF THE DEFENCE (GENERAL) REGULATIONS, 1939.

THIS Order specifies further conditions for the purchase or sale of sulphate of ammonia.

Department of Scientific and Industrial Research

METHODS OF ANALYSIS OF COAL AND COKE‡

THIS Paper supersedes Survey Paper No. 7, which was published in 1927. It embodies the results of further investigation of details of the earlier methods, and includes methods for the examination of coke. The principal changes in the existing methods relate to those for the determination of volatile matter, nitrogen, phosphorus, and agglutinating value; methods included for the first time are for the determination of chlorine, arsenic and the forms of sulphur; additional methods are given for the determination of carbon dioxide and sulphur; and the section on the Gray-King carbonisation assay, previously dealing only with the low-temperature form, now includes a description of the apparatus and method for the high-temperature modification (at 900° C.), which is intended for the rapid evaluation of coals for the carbonisation industries.

VOLATILE MATTER.—The standard method described (heating for 7 minutes at 925° C.) should be used for all coals that give a coherent coke button and do not decrepitate. A modification, in which a slower rate of heating is employed, is used for coals that decrepitate, and another modification (admixture with non-caking coal) to prevent losses due to entrainment and decrepitation. An alternative method, requiring a more elaborate apparatus, may be used for anthracites and non-caking coals containing up to 12 per cent. of volatile matter. For coal the maximum furnace temperature should be 925° C., whilst for coke 950° C. is specified.

NITROGEN.—The method specified is a modification of the Kjeldahl process in which selenium is used as catalyst. The use of mixtures of selenium and mercury compounds is not recommended, for, although there is a slight saving in time during the combustion, subsequent addition of sodium sulphide becomes necessary.

PHOSPHORUS.—In the method previously specified several fusions of the ash with sodium carbonate were required, with extraction of each melt with water until all phosphorus had been removed. The new method consists in removing silicon from the ash by evaporation with mixed hydrofluoric and nitric acids, precipitating the phosphorus as phosphomolybdate, and titrating the precipitate. It was found by the Northern Coke Research Committee that comparatively

* H.M. Stationery Office, York House, Kingsway, London, W.C.2. Price 1d. net.

^a S.R. & O., 1940, No. 1712.

† H.M. Stationery Office. Price 1d. net.

‡ Fuel Research Paper No. 44. Physical and Chemical Survey of the National Coal Resources, October 28th, 1940. H.M. Stationery Office, York House, Kingsway, London, W.C.2. Price 2s. net.

excessive amounts of arsenic or titanium did not affect the results materially, but a special procedure may be necessary for ashes containing much vanadium.

AGGLUTINATING VALUE.—In the empirical test described, the agglutinating value is the maximum whole-number ratio of sand to unit coal in a mixture of sand and coal which, after carbonisation under specified conditions, gives a coherent mass capable of supporting a 500-g. weight, with less than 5 per cent. of the mixture remaining as a loose powder.

CHLORINE.—No practical method for the determination of water-soluble chlorine can be recommended. For the determination of total chlorine the coal is mixed with anhydrous sodium carbonate, and the mixture is heated at $475^{\circ} \pm 25^{\circ} \text{C.}$, in a muffle furnace until combustion is complete (about 20 hours). The chlorine is extracted with dilute nitric acid, the solution is treated with excess of standard silver nitrate solution, and the excess of silver nitrate is titrated with potassium or ammonium thiocyanate solution.

The amount of chlorine in coal usually ranges from 0.05 to 0.20 per cent., but some coals may contain as much as 1.0 per cent. and others as little as 0.01 per cent.

ARSENIC.—Arsenic is present in most coals, but usually in traces—less than 3 or 4 p.p.m. (as arsenious oxide), though in exceptional instances 40 to 50 p.p.m. may occur. In the method described the coal (ground to pass a 72-mesh B.S. sieve) is incinerated with a mixture of magnesium oxide, sodium carbonate and potassium nitrate, similar to that proposed by Hertzog (*Ind. Eng. Chem., Anal. Ed.*, 1935, 7, 163). The bases retain the arsenic, whilst the potassium nitrate maintains oxidising conditions and prevents the arsenic compounds being reduced to more readily volatilised arsenious compounds. After the incineration the arsenates are reduced to arsenites by means of sulphur dioxide, to ensure the subsequent complete reduction to arsine.

The arsenic is determined by a modification of the Gutzeit method, the arsenic being reduced to arsine, which is brought into contact with a paper strip impregnated with mercuric bromide. The length of the stain formed on the strip is a measure of the amount of arsenic. The apparatus described and illustrated is a modification of that devised by Crossley (*J. Soc. Chem. Ind.*, 1936, 55, 272r). The bottom section of the delivery tube contains filter-paper impregnated with lead acetate, and the middle section is packed with glass wool moistened with lead acetate solution. Any hydrogen sulphide is removed in these sections, whilst the arsine passes on to the mercuric bromide paper in the top section.

SULPHUR.—"Sulphate" sulphur is determined by extracting the coal with dilute hydrochloric acid and determining the sulphur in the extract. "Pyritic" sulphur is insoluble in dilute hydrochloric acid, but is quantitatively dissolved by dilute nitric acid. The "pyritic" iron is determined in the nitric acid extract, and the amount of sulphur corresponding to FeS_2 is calculated from the result. The two acid extractions may be carried out on the same portion of the coal sample. If they are made simultaneously, it is necessary to determine the iron in the hydrochloric acid extract to obtain the correction for the "pyritic" iron determination.

REPRODUCIBILITY OF ANALYTICAL RESULTS.—The degree of divergence to be expected between determinations made in the same laboratory or in different laboratories is discussed in an appendix. Only for volatile matter was the divergence in experimental results large enough to be serious. This is attributed partly to the fact that the apparatus used had not been fully standardised in different laboratories. Another appendix deals with the methods of reporting results, and particularly the method of calculating them on the basis of mineral-free matter or pure coal substance.

New South Wales

ANNUAL REPORT OF THE GOVERNMENT ANALYST FOR THE YEAR 1938

THE total number of samples examined in the Chemical Laboratory is reported by Mr. S. G. Walton (Government Analyst) to be 33,655, including 31,079 under the Pure Food and Milk Acts, and 2576 for the Public Services. Of the 21,947 samples of milk examined, 1.36 per cent. of those from the metropolitan area were adulterated and 7.23 per cent. of the country samples.

BROWN BREAD.—The trade asked that the standard for fibre content should be lowered from the present figure (1.2 per cent.) to 0.75 per cent. As this would be equivalent to permitting brown bread to be made from a mixture of about 1 part of whole wheat flour with 2 parts of white flour, it was decided not to alter the present standard.

WHOLEMEAL BREAD.—Deficiencies in the crude fibre content of wholemeal bread led to several prosecutions during the year. In fixing the standard a generous tolerance was allowed, the minimum figure (2 per cent.) being less than that usually found. Thus the proportion of crude fibre in wholemeal milled in the Department of Agriculture ranged from 2.38 to 2.55 per

cent. Analyses were also made of wholemeal breads, prepared and baked in the Department of Agriculture, and the results were as follows:

| Proportion of wholemeal used Per Cent. | Maximum moisture Per Cent. | Crude fibre (calculated on dry substance) | |
|---|-------------------------------|---|-------------------------|
| | | Determined Per Cent. | Calculated Per Cent. |
| 100 | 45.7 | 2.57 | 2.57 |
| 80 | 45.4 | 2.12 | 2.06 |
| 60 | 45.0 | 1.66 | 1.54 |
| 40 | 45.0 | 1.12 | 1.03 |
| 100 + 5 per cent. of gluten | 46.4 | 2.50 | 2.45 |

BAKING POWDER AND SELF-RAISING FLOUR.—A large manufacturing company requested the deletion of the Regulation requiring the declaration, on the label of packages containing self-raising flour and baking powder, of the presence of acid phosphate, where either sodium or calcium phosphate was used as the acid constituent of the aerating medium. An investigation of the baking and keeping properties of different types of self-raising flour and baking powder was therefore undertaken. The acid constituent of the samples prepared was (1) cream of tartar, (2) cream of tartar and tartaric acid, (3) calcium acid phosphate, and (4) sodium acid pyrophosphate. A self-raising flour was also prepared containing a mixture of calcium acid phosphate and sodium acid pyrophosphate (No. 6), a cream of tartar self-raising flour (No. 5) being used for comparison. The samples of baking powders and self-raising flours were prepared to give a theoretical yield of 15 per cent. by weight, and 52 grains per lb., respectively, of available carbon dioxide. In the tartrate powders and flours the acid ingredient and sodium bicarbonate were mixed to give an exact chemical balance, whilst the acid phosphate preparations were made according to formulae supplied, the acid ingredient being in slight excess. The actual available carbon dioxide of each sample was determined at the time of making the baking tests. Each powder and flour was stored under normal conditions for about 3 months, to ascertain its keeping qualities, the baking powders being packed in lever-lid tins and the flours in paper one side of which was glazed. After 3 months' storage the available carbon dioxide was again determined. The following percentage results, showing (a) original available carbon dioxide and (b) available carbon dioxide after 3 months, were obtained:—No. 1 (a) 14.58, (b) 13.09; No. 2 (a) 14.74, (b) 13.09; No. 3 (a) 11.9, (b) 4.68; No. 4 (a) 14.3, (b) 13.82.

These and other experiments showed that powders and flours prepared with acid calcium phosphate were inferior to tartrate preparations in keeping properties, but that the latter were inferior to acid sodium pyrophosphate preparations.

The baking tests were made by an expert at the East Sydney Technical College, scones and sponges being baked under standardised conditions. For the scones the grading of the powders by the expert in order of merit was 1, 2, 4, 3 and for the sponges it was 1, 2, 3, 4. In every instance the tartrate baking powder and self-raising flour yielded products superior in texture, flavour and volume. The articles baked with acid phosphate invariably had a marked flavour to which the Australian palate would have to become accustomed.

As a result of this investigation it was decided not to accede to the request mentioned above, but permission was given to alter the declaration required by deletion of the word "acid."

STANDARDS FOR CHEESE.—The Department of Agriculture concurred in alterations recommended, and these were given effect to by the Pure Food Advisory Committee. The revised standards are as follows:

- (1) *Cheese* is the solid or semi-solid product obtained by coagulating milk, cream or skim-milk with rennet or acid. It may contain ripening ferments, seasonings, salt (sodium chloride), flavourings, and permitted colouring matter. It shall not contain any foreign fat. For the purpose of this Regulation, milk shall be deemed to be the milk of any domestic animal.
- (2) *Cream cheese* shall contain not less than 70 per cent. of milk-fat in its water-free substance, and not more than 50 per cent. of water.
- (3) *Full or whole milk cheese* shall contain not less than 50 per cent. of milk-fat in its water-free substance, and not more than 40 per cent. of water.
- (4) *Skim-milk cheese* is cheese which contains less than 30 per cent. of milk-fat in its water-free substance.

There shall be written in the statement or label attached to every packet which contains skim-milk cheese or on the cheese itself in bold-faced sans-serif capital letters of not less than 18 points face measurement in such colours as to afford a distinct colour contrast to the ground, the words "Skim-milk Cheese."

- (5) Cheese sold under the name of Edam, Gruyère, Gouda, Stilton, or any other generally known name, shall correspond thereto in respect of composition and character.
- (6) Cheese sold without name or classification shall be deemed to be full or whole-milk cheese, and shall comply with the standard prescribed therefor.
- (7) (a) *Processed cheese, cheese paste or cheese spread*, not otherwise named to indicate the variety of cheese, shall contain not less than 45 per cent. of milk-fat in its water-free substance,

not more than 45 per cent. of water, and not more than 3 per cent. of harmless emulsifying agents.

- (b) Every packet of processed cheese shall bear a label in which shall be written in bold-faced sans-serif capital letters of not less than 12 points face measurement in such colours as to form a distinct colour contrast to the ground, the word "Processed."
- (8) *Luncheon cheese, cheese mixture or club cheese* is a cheese compound prepared from cheese with or without wholesome foodstuffs and condiments, and shall contain not less than 48 per cent. of milk-fat in its water-free substance, and not more than 35 per cent. of water.
- (9) The addition to luncheon cheese, cheese mixture or club cheese of sulphur dioxide (or sulphites calculated as sulphur dioxide) in proportion not exceeding 2 grains to the pound is hereby permitted.

d-SORBITOL IN DIABETIC CHOCOLATE.—An application was received for permission to use *d*-sorbitol in place of sucrose in the manufacture of diabetic chocolate. In view of the fact that its use is allowed by the existing standard for invalids' foods, permission was given. The addition of 3 grains of saccharin per lb. was also permitted.

ALKALINE CREAM.—A large number of samples of cream brought into New South Wales from an adjoining State were found to contain an alkaline preservative, probably sodium peroxide. After successful prosecutions samples of cream no longer gave reactions for peroxide, but still contained a foreign alkaline substance either used as a neutralising agent or being the residue from a preserving process. This addition was detected by determining the pH value, and legal proceedings were taken whenever the pH exceeded 7. The recorded values for the pH of cows' milk show a considerable range (6.4 to 7.2). The normal range given by Van Slyke and Barker (*J. Biol. Chem.*, 1919, 345) is 6.5 to 6.75. It is unlikely therefore that the pH of any commercial cream would exceed 6.85 unless alkaline neutralisers had been used. Fifteen samples of cream found to contain alkaline peroxide had pH values ranging from 7.20 to 7.50, whilst 8 samples, reported to contain an alkaline neutraliser but no peroxide, had pH 7.18 to 7.45. Later samples showed values less than 7.0, and in some of these it is probable that the alkalinity from the peroxide process had been balanced by an addition of acid. A low freezing-point (Hortvet) (−0.57 to −0.58) supported this conclusion.

NITROUS OXIDE FOR WHIPPED CREAM.—An application was received for permission to use nitrous oxide in whipped cream. The application was not recommended because the introduction was for the purpose of increasing the bulk, and also in view of the absence of definite information as to the harmlessness or otherwise of nitrous oxide.

DIETHYLENE GLYCOL IN ESSENCE OF LEMON.—Two samples of essence of lemon contained diethylene glycol as the vehicle. As this substance cannot be considered harmless, its use as a solvent in essences is a contravention of the standard under the Act.

STANDARD FOR MALTED MILK.—The following standard, based on the results of the examination of all the brands of malted milk sold in New South Wales, has been adopted as reasonable for both the manufacturer and consumer.

- (1) Malted milk powder is the product made either
- (a) by combining whole milk with the liquid separated from a mash of ground malt and wheat or other cereal flour in such a manner as to secure full enzymic action of the malt extract and reducing the product to dryness by desiccation under reduced pressure; or
 - (b) by mixing dried whole milk with dried malt extract.
- (2) Malted milk powder shall contain not less than 7.5 per cent. of milk-fat, not more than 3.5 per cent. of water, and may contain salt (sodium chloride) not exceeding 1 per cent. It shall be free from rancidity.
- (3) Malted milk for retail sale shall be packed in airtight receptacles.

STANDARD FOR TRIPE.—With a view to ensuring that a uniform article shall be supplied, the following revised standard has been adopted:

Tripe as sold for human consumption shall not be prepared so as to impair its nutritive properties, nor contain any added foreign matter except salts, and its reaction value (determined on the liquid obtained by exudation and pressure) shall not be less than pH 6.5 nor greater than pH 7.5.

ZINC IN CANNED VEGETABLES.—The deep green colour of certain vegetables was found to be due to the presence of organically combined zinc. Successful prosecutions caused the manufacturers to abandon the use of zinc.

Other samples of canned vegetables were found to be coloured with an aniline dye; this practice was also stopped by legal proceedings. Subsequently a request for permission to use an aniline dye for this purpose was received, but after consideration permission was refused.

Patiala (Punjab) India

REPORT OF THE CHIEF CHEMIST FOR THE YEAR ENDED APRIL 12TH, 1940

THE Central Analytical Laboratory of the State of Patiala was established in March, 1939, at the Mahendra College, under the direction of Mr. K. L. Budhiraja, M.Sc., A.I.C. It was set up primarily to exercise control over the Ghee Grading Station at Namaul, and to analyse soils and minerals from the Development Department, but in practice it has been doing work for all the State Departments. The 307 samples submitted consisted mainly of ghee, but there were also 69 samples of soils, 11 samples of minerals, and 16 samples of water. When the Pure Food Act comes into force the work of the Laboratory is certain to expand.

GHEE.—Of the 211 samples submitted, 83 were received from the Ghee Grading Station, 73 from executive authorities, and the remainder from Medical Officers of Health, etc. In the absence of any standards for purity, the Patiala State adopted those laid down by the Agricultural Marketing Adviser to the Government of India. Fifty-five of the samples (other than those from the Grading Station) were genuine, 69 were adulterated, and the others were of doubtful purity. The degree of adulteration ranged from 5 to 70 per cent., and 15 samples consisted of pure hydrogenated vegetable fat popularly called vanaspati or vegetable ghee; this is the most common adulterant. Prosecutions were undertaken and fines were imposed as the result of analyses at the Laboratory. The Department was also helpful in checking the smuggling of vegetable ghee products in evasion of the local (octroi) duties.

Ceylon

ANNUAL REPORT OF THE GOVERNMENT ANALYST FOR THE YEAR 1939

THE Government Analyst (Mr. J. V. Collins) reports that the war has not affected the analytical work of the Department, and that there has been a small increase in the number of articles examined (4255 as compared with 4095 in 1938). Much of the work is concerned with criminal investigation; 1091 cases were investigated as against 1041 in 1938.

STAIN CASES.—There were 128 cases in which identification of blood was required, and 150 cases (with 323 exhibits) in which identification of the species of blood was asked for. Of these exhibits, 195 gave positive reactions for human blood and 20 for blood other than human.

An interesting case was one in which a heavy blood stain on the vest of an accused person was found, very unexpectedly, to be a mixture of ox blood and pig blood. On inquiry it was found that the accused had been arrested by a public-spirited butcher, who had knocked him down with the first weapon that came to hand—a joint of beef. This joint was on the butcher's stall, and above it was suspended a joint of pork.

POISONING CASES.—Exhibits from the Courts were examined in 84 cases, and poison was detected in 46. The poisons identified included copper salts (7), powdered glass (5), arsenic (4), arsenic and mercury (2), nitric acid and nitrates (3), prussic acid (3), disinfectants (3), croton oil (3), quinine (2), nux vomica (2), dyestuffs (2), mydriatic alkaloids (2), acetic acid (2), bleaching powder, a saponin, an unidentified alkaloid, etc.

Copper salts are becoming increasingly popular, owing apparently to a misguided belief in their efficacy in abortion cases. The dyestuffs are probably used as "charms" or are added mischievously, as the usual history is that some one introduced an unknown substance into a pot of cooking rice, which subsequently became brightly coloured.

Quinine Poisoning.—In the cases of quinine poisoning two hospital patients received an overdose of quinine hydrochloride amounting to nearly 100 grains. Both patients died shortly after, and fits were a symptom in each case. The following quantities of anhydrous quinine (in grains) were recovered:—Stomach and contents, 10, 11; kidneys, $1/3$, $5/13$; liver, $1\frac{1}{3}$, 1; brain, trace in each case.

DECIPHERING NUMBERS ON BICYCLES.—Sixty bicycles, as compared with 3 in 1938, were examined to discover the original number. There was no evidence of alteration of the number on 16 of these; on 10 it was not possible to restore the original number; some of the original figures could be made out on 9, and on 25 the whole of the original number could be deciphered.

MILK.—Of the 659 samples examined, 391 were adulterated. It is remarkable that for many years past a higher percentage of genuine samples has been received from Local Authorities than from the Medical Department—46 per cent. as against 32 per cent. Cane sugar was found in 20 samples, mostly of milk supplied on contract to Government hospitals. Turpentine, an unusual constituent, was found in one sample.

ARRACK.—In connection with the control of Government arrack, 124 samples from distilleries and warehouses were examined for the Excise Department. As a result of cleaner collecting and better fermentation of the toddy, the quality of the spirit is improving yearly and the acidity figure is falling to a more satisfactory level.

Standardisation of Testing Methods for Textiles

DEFINITIONS OF ATMOSPHERIC HUMIDITY AND THE MOISTURE CONTENT AND REGAIN OF TEXTILE MATERIALS*

THE following definitions are given for reference in connection with Standard Specifications. Standardised methods for determining the physical constants here defined are to be published later.

ABSOLUTE HUMIDITY AND RELATIVE HUMIDITY.—The absolute humidity of the atmosphere is defined as the mass of water present per unit volume. The relative humidity is the ratio of the actual amount of water per unit volume to the amount necessary to saturate a unit volume at the same temperature and pressure. The relative humidity is usually expressed as a percentage.

DEW POINT.—If moist air is cooled, condensation occurs as the state of saturation is approached. The temperature at which condensation begins is termed the dew point.

MOISTURE-CONTENT.—The percentage moisture-content means the amount of moisture in a material when expressed as a percentage of the total weight.

REGAIN OF TEXTILE MATERIALS.—When the moisture in a textile material is calculated as a percentage on the oven-dry weight, the percentage is termed the regain. Thus, with a moisture-content of 20 per cent., the regain is $100 \times 20 \div 80$, i.e. 25 per cent.

Standard Regains.—These are arbitrary values chosen as the most useful for commercial purposes. The correct conditioned or "correct invoice weight" of a consignment shall be calculated from its oven-dry weight and the recognised standard regain.

More or less arbitrary percentage moisture-contents and regains have been attributed to the different textile fibres and the yarns and cloth made from them, so that for trade purposes they may be declared "in correct condition" when they contain the recognised percentages of moisture or the equivalent "regain." The following "standards of regain" are commonly, but not universally, accepted:

| | Standard moisture-content Per Cent. | Standard regain Per Cent. |
|---|---|---------------------------------|
| Cotton | 7.83 | 8.5 |
| Silk | 9.91 | 11 |
| Flax and hemp | 10.71 | 12 |
| Jute | 12.00 | 13.75 |
| Wool and waste | 13.79 | 16 |
| Wool (tops combed with oil) | 15.97 | 19 |
| Wool (tops combed without oil) | 15.43 | 18.25 |
| Worsted yarns | 15.43 | 18.25 |
| Carded woollen yarns | 14.53 | 17 |
| Wool noils (ordinary) | 12.28 | 14 |
| Wool noils (scoured and carbonised) | 13.79 | 16 |
| Woollen and worsted cloths | 13.79 | 16 |
| *Viscose and cuprammonium rayons | 9.91 | 11 |
| *Cellulose acetate rayon | 5.66 | 6 |

* Other standards are applicable in certain circumstances.

When these standards were first made, it was assumed that when 100 parts of the fibre in the oven-dry condition were exposed to the "usual condition of the atmosphere" they would "regain" moisture to the extent shown in the foregoing table. When the moisture-contents and the regains of the material agree with the standards in the table, they are regarded as "100 per cent. in correct condition."

Equilibrium Regain.—After exposure for a sufficient time to an atmosphere whose relative humidity and temperature are kept constant, the regain of the material reaches a steady value termed its "equilibrium regain."

STANDARD ATMOSPHERE.—Before mechanical or physical tests are applied, textile materials must be "conditioned" by exposure under standard conditions of relative humidity and temperature. In this country and in the United States the conditions denoted by 65 ± 1 per cent. relative humidity at $70^\circ \pm 2^\circ$ F. are being widely observed. On the Continent the corresponding standards are $65, \pm 1$ per cent. and $20^\circ \pm \frac{5}{2}^\circ$ C., the greater tolerance for temperature being allowed to avoid, as far as possible, refrigeration in the summer.

DETERMINATION OF RELATIVE HUMIDITY*

Under normal conditions the ventilated wet and dry bulb hygrometer and the sling hygrometer give results of sufficient accuracy for commercial purposes. Other methods, capable of greater accuracy, depend on the direct determination of the dew point or the weighing of the moisture in a measured quantity of air.

Various empirical relations connecting the wet bulb and dry bulb temperatures with the dew point have been derived and tables have been computed to avoid the calculation of the relative humidity. Glaisher's *Hygrometric Tables* are no longer used in scientific work. For the ventilated hygrometer and the sling hygrometer the psychrometric tables issued by the Russian Meteorological Institute have been amplified and are widely used at the National Physical Laboratory, Teddington. The tables in the Appendix to the Standards on Textile Materials prepared by Committee D13 of the American Society for Testing Textile Materials are based on the Smithsonian Meteorological Tables. Marvin's *Psychrometric Tables*, published by the U.S.A. Dept. of Agriculture, are used fairly generally; they can be obtained from Casella & Co., Ltd., Fitzroy Square, London, W.1.

In other instruments there is a sensitive material, such as human hair or goldbeaters' skin, and the changes in this resulting from the absorption or loss of water are recorded. These instruments should not be used in cases of dispute and should be calibrated regularly by comparison with a standard hygrometer.

British Standards Institution

BRITISH STANDARD No. 598—1940. METHODS FOR THE SAMPLING AND EXAMINATION OF BITUMINOUS ROAD MIXTURES†

THIS Specification, first issued in 1936, has recently been revised, and several modifications have been introduced to keep it up-to-date with present practice. In addition to the complete revision of certain sections, a modified test for the rapid determination of bitumen-content and two alternative methods for the recovery of bitumen have been introduced.

The rapid method for the determination of soluble bitumen is that described in the *J. Inst. Petroleum*, 1939, 25, 185, 168–177, but in cases of dispute the standard method, which is essentially that described in *J. Soc. Chem. Ind.*, 1931, 50 (*Chem. and Ind.*), 599, must be used.

The two alternative methods for the recovery of soluble bitumen are to be regarded as tentative until experience in their use has been gained.

DEFINITIONS.—The following definitions, which (with the exception of that for asphalt) have been adopted by the Permanent International Association of Road Congresses, are applicable throughout.

Bitumen.—Mixtures of hydrocarbons of natural or pyrogenous origin or combinations of both (frequently accompanied by their non-metallic derivatives) which can be gaseous, liquid, semi-liquid or solid, and which are completely soluble in carbon disulphide.

Asphaltic Bitumen.—Natural or naturally occurring bitumen, or bitumen prepared from natural hydrocarbons by distillation or oxidation or cracking; solid or viscous, containing a low proportion of volatile products; possessing characteristic agglomerating properties, and substantially soluble in carbon disulphide.

Asphalt.—A natural or mechanical mixture in which asphaltic bitumen is associated with inert mineral matter. An indication of the origin or nature of "asphalt" should always be given.

Tar.—A bituminous product, viscous or liquid, resulting from the destructive distillation of organic material. The word "tar" should be preceded by the name of the substance from which it was derived, and its mode of production should also be indicated.

CONTENTS OF THE SPECIFICATION.—Part I, Definitions. Part II, Sampling: (1) Asphalt, (2) Tarmacadam, (3) Information to be supplied with sample. Part III, Methods of Examination: Pot stain test—Apparent specific gravity—Voids—Soluble bitumen-content: (1) Hot and cold asphalt, (2) Tentative method for recovery of bitumen, (3) Tarmacadam.—Water-content: (1) Asphalt, (2) Tarmacadam.—Mineral aggregate: (1) Tests on mineral aggregate, (2) Preparation of sample, (3) Petrographical identification—Certificate of analysis—Appendix: Method for the rapid determination of soluble bitumen.

* *J. Text. Inst.*, 1940, 31, 55.

† Copies of the Specification can be obtained from the British Standards Institution, 28, Victoria Street, London, S.W.1. Price 2s.; post free 2s. 3d.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Simplified Alkali-liability Determination for Starch Products. T. J. Schoch and C. C. Jensen. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 531-532.)—Raw starch decomposes in hot aqueous alkali to give simple acidic substances, principally formic, acetic and lactic acids as well as pyruvic aldehyde, and this suggests that the reaction is initiated by enolisation of free terminal aldehyde groups as formulated by Evan (*J. Amer. Chem. Soc.*, 1926, 48, 2665; 1930, 52, 294) for the aldose sugars. With acid-modified starches alkaline decomposition proceeds more rapidly, indicating increased aldehyde-content. The reaction provides a simple and precise measurement of alkali-liability, the rate of decomposition of the starch (the alkali number) being the number of ml. of 0.1 N sodium hydroxide solution consumed by each g. of starch during digestion with alkali at 100° C. for one hour. The starch should be pulverised to pass through a 60-mesh sieve, and its moisture-content determined separately by drying *in vacuo* at 105° C. for 4 hours, the alkali number being calculated for the dry substance. When a starch product contains sufficient added acid or alkali to affect the alkali number, 1 g. is gelatinised in hot water and neutralised to thymol blue with standard acid or alkali, and the alkali number is corrected for this titre. The digestion with alkali may conveniently be made in a feeding bottle provided with its rubber cap which is pierced by means of a hot needle to allow an exit for steam. The starch (500 mg.) is gently agitated with 10 ml. of water to form a uniform suspension, 25 ml. of 0.4 N sodium hydroxide solution are added with continued agitation and finally 65 ml. of hot water, and the bottle is capped and placed in a vigorously boiling water-bath. If the starch product gelatinises in cold water, it is first treated in a dry bottle with 1 or 2 ml. of benzene before addition of the alkali and water. The bottle is heated for

exactly an hour, after which it is placed in cold water, and 50 to 75 ml. of water are added to stop the decomposition process. The excess alkali is titrated to the yellow end-point of thymol blue with standard acid and, as a blank determination, 25 ml. of the alkali solution are titrated with the standard acid. Strict adherence to the conditions described is recommended. The method has been applied to a number of theoretical and practical problems. Commercial maize and wheat starches have higher alkali numbers than the tuber starches, and this is attributed to different molecular configuration rather than to hydrolytic decomposition during manufacture. A particular starch is not necessarily composed of identical molecules. By leaching maize starch at a temperature just below gelatinising point by the method of Baldwin (*J. Amer. Chem. Soc.*, 1930, 52, 2907) a small amount of soluble carbohydrate with a high alkali number and probably consisting of small molecules was obtained, and a typical acid-modified thin-boiling starch exhibited a similar heterogeneity. When raw maize starch is digested with hot alkali and the neutralised solution is treated with alcohol the precipitated carbohydrate has an alkali number lower than that of the original starch, but even when the pre-digestion with alkali is continued until nearly all the starch is destroyed the alkali number of the residual carbohydrate does not fall below 4.0. With increasing degree of acid conversion, the alkali number rises progressively and reaches values from 56 to 66 for the white dextrins and Lintner's soluble starch. When white dextrin is dried at 105° C. there is a progressive fall in alkali-liability, and this may be due to the formation of anhydrides less susceptible to attack by alkali. Alkali-liability must not be regarded as a quantitative evaluation of aldehyde-content but merely as an empirical index of the degree of hydrolysis.

A. O. J.

Component Acids and Glycerides of some Indian Ox Depot Fats. T. P. Hilditch and K. S. Murti. (*Biochem. J.*, 1940, 34, 1301-1311.)—The analytical charac-

teristics of four different Indian ox depot fats (taken mainly from the perinephric tissues) were as follows:

| Fat from | Sap. val. | Iodine val. | Free fatty acids (as oleic) Per Cent. | Unsap. matter Per Cent. | Fat, m.p. (open tube) ° C. | Mixed acids, setting-point ° C. |
|---------------------------------------|-----------|-------------|---------------------------------------|-------------------------|----------------------------|---------------------------------|
| Bombay cow, original .. | 280.1 | 26.6 | 1.4 | — | — | — |
| " " neutralised .. | 282.2 | 26.5 | 0.1 | 0.35 | 50.5 | 47.6 |
| " bullock, original .. | 282.0 | 26.1 | 2.4 | — | — | — |
| " " neutralised .. | 283.2 | 25.8 | 0.1 | 0.3 | 50.0 | 48.5 |
| Calcutta cow and bullock, original .. | 283.2 | 31.0 | 1.3 | 0.2 | 49.0 | 48.4 |
| Calicut cow, original .. | 283.5 | 31.1 | 0.2 | 0.4 | 50.5 | 48.7 |

From each fat, neutralised, if necessary, the mixed fatty acids were liberated and separated by the usual lead salt and alcohol method; the methyl esters of the respective acids were

then fractionally distilled. The mixed acids were found to possess the following composition (in mols. per cent.):

| | Bombay cow | Bombay bullock | Calcutta ox and bullock | Calicut cow |
|--------------------------------------|------------|----------------|-------------------------|-------------|
| Saturated acids: | | | | |
| Lauric | 0.2 | 0.3 | 0.3 | 0.5 |
| Myristic | 5.2 | 4.4 | 2.8 | 3.7 |
| Palmitic | 43.3 | 39.1 | 38.8 | 34.7 |
| Stearic | 22.9 | 27.9 | 25.5 | 27.9 |
| Arachidic | 0.4 | 1.0 | 0.4 | — |
| Unsaturated acids: | | | | |
| Tetradecenoic | 0.5 | 0.5 | 0.4 | 0.4 |
| Hexadecenoic | 1.4 | 1.0 | 2.2 | 1.6 |
| Oleic | 25.1 | 24.8 | 28.0 | 29.5 |
| Octadecadienoic | 0.9 | 0.9 | 0.9 | 1.3 |
| C ₂₀₋₂₂ unsaturated | 0.1 | 0.1 | 0.7 | 0.4 |

Compared with the composition of English, North and South American and Australian ox depot fats previously examined, the unsaturated acid content (27 to 33 per cent.) is low, the acids from other ox depot fats never containing less than 40 per cent. and frequently as much as 50 per cent. The deficiency of oleic acid in the Indian fats is made up by an increased proportion not of stearic acid, but of palmitic acid, and the proportion of this acid is indeed higher than has hitherto been observed in any animal depot fat. For a wide range of land animals (including birds) the palmitic acid content has been found to be 25 to 30 per cent. As a consequence of the high proportion of palmitic acid the

"melting-points" of the fats and the setting-points of their mixed fatty acids are little higher than those of the usual ox depot fats; a high proportion of stearic acid, on the other hand, would have resulted in a considerably higher melting-point. The Bombay cow and Calicut cow depot fats were further examined to determine the nature of their component glycerides. The fats were resolved by systematic crystallisation from acetone into three fractions of different consistence and iodine value, and these were separately examined as previously described (Hilditch and Paul, *Biochem. J.*, 1938, **32**, 1775). The probable component glycerides (mols. per cent.) are as follows:

| | Bombay cow | Calicut cow |
|--------------------------------|--------------|--------------|
| Fully saturated: | | |
| Tripalmitin | 3.0 | — |
| Dipalmitostearin | 22.6 | 16.5 |
| Palmitodistearin | 10.3 | 11.8 |
| Mono-"oleo"-disaturated: | | |
| Hexadecenodistearin | — | 0.1 |
| Hexadecenopalmitostearin | 2.4 | — |
| "Oleo"-dipalmitin | 18.2 | 11.0 |
| "Oleo"-palmitostearin | 33.0 to 32.2 | 41.5 to 38.4 |
| "Oleo"-distearin | — | 1.8 |
| Di-"oleo"-monosaturated: | | |
| Palmitodi-"oleic" | 9.7 to 10.5 | 14.2 to 17.3 |
| Stearodi-"oleic" | 0 to 0.8 | 0 to 3.1 |
| Tri-"oleic" | 0.8 to 0 | 3.1 to 0 |

The glycerides of the Indian ox depot fat are thus of the mixed type characteristic of all other animal depot fats, but the Calicut fat shows a much closer resemblance to an English fat than does the Bombay fat; the total of fully saturated components in the Calicut fat (28 per cent.) is, however, higher

than in English fat (17 per cent.). The Bombay cow depot fat is remarkable in possessing the highest proportion of fully saturated glycerides (36 per cent.) yet observed in an animal depot fat; the proportions of dipalmitostearin and of "oleo"-dipalmitin are abnormally high. F. A. R.

Mixed Unsaturated Glycerides of Liquid Seed Fats. II. Low Temperature Crystallisation of Cottonseed Oil. T. P. Hilditch and L. Maddison. (*J. Soc. Chem. Ind.*, 1940, **59**, 162-168; cf. *ANALYST*, 1940, **65**, 364.)—The method of crystallisation of solid fats from acetone at 0°C. or -10°C., has been extended to cottonseed oil as representing the more liquid and unsaturated fats, but much

lower temperatures have been used. The component acids (per cent. by weight) of the cottonseed oil were: myristic, 1.4; palmitic, 23.4; stearic, 1.1; arachidic, 1.3; tetradecenoic, 0.1; hexadecenoic, 2.0; oleic, 22.9; linolic, 47.8. The oil was separated into 6 fractions by systematic crystallisation from acetone at temperatures down to -35°C., and the acids in each fraction were determined. The

technique was simplified by adding solid carbon dioxide directly to the acetone solution in the following manner:—The acetone solution of the fat was placed in a 3-litre round-bottomed flask contained in an insulated rectangular cardboard box, which had a movable fibre partition and was fitted with a similar lid, circular holes being cut in each to take the neck of the flask. At the bottom of the box there was a layer of three inches of cotton wool. The fraction of the cotton-seed oil to be crystallised was dissolved in dry acetone (3 to 7 ml. per g. of fat) in the flask, and small pieces of solid carbon dioxide (not larger than $\frac{1}{4}$ -inch cubes, wiped free from adherent frost) were added to the solution, with vigorous stirring, further portions being added when the evolution of carbon dioxide gas ceased, until the temperature had fallen to the desired point. The space between the movable partition (resting on the shoulder of the flask) and the lid of the box was then loosely packed with larger lumps (about 1-in. cubes) of solid carbon dioxide. In this way the solution could be held for any desired period (e.g. up to 7 hours) at a temperature varying by not more than $\pm 2^\circ \text{C}$., but thorough stirring at intervals was necessary to prevent local over-cooling. The separated fraction was removed by filtration with the aid of a pump, the low temperature being maintained by insulating the Buchner funnel in a box packed with crushed solid carbon dioxide. The stem of the funnel passed through a hole in the base of the box and was connected with a large filter-flask which was not artificially cooled. The solid fat fraction collected in the funnel was washed with 100–200 ml. of acetone previously cooled with solid carbon dioxide to -45°C ., and was then heated at 100°C . *in vacuo* to remove all acetone, whilst the acetone filtrates were distilled and the residual dissolved fat was also heated at 100°C . *in vacuo*.

The possible component glycerides in each fraction thus obtained were then considered in the light of the component acids found, but without the help which with the more saturated fats was to be had from a study of the fully saturated components present, or of the products of hydrogenation of the fractions. Sufficient separation was effected to show that each fraction was to a very large degree made up of only 2 of the 4 possible categories of mixed saturated unsaturated glycerides—fully saturated, mono-unsaturated-disaturated, di-unsaturated-monosaturated and tri-unsaturated glycerides. Large proportions of mixed oleolinoleo-glycerides and extremely small proportions of the simple forms, trilinolein or triolein, appeared to be present in each fraction. Conclusive quantitative data for the component glycerides in each fraction could not be obtained from the component acid analyses alone, but the following conclusions were reached:—The chief glycerides in the cotton-seed oil examined were about 58 per cent. of saturated (mainly palmito-) di-unsaturated glycerides, accompanied by about 28 per cent.

of tri-unsaturated glycerides; smaller proportions (18 per cent.) of mono-unsaturated-disaturated glycerides, and very small traces of palmitin. Of the main components, 35–40 per cent., were probably palmito-oleolinoleins, with 20 per cent., or somewhat more, of palmito-dilinoleins and possibly small amounts of palmito-diolein; oleodilinoleins formed probably almost the whole of the tri-unsaturated glycerides, although here again the presence of small amounts of triolein was not excluded. The minor quantities of mono-unsaturated glycerides were made up of somewhat more linoleo- than oleo-compounds, and the saturated acyl radicals present quite possibly included one palmitic and one minor component saturated acid (myristic, stearic or arachidic) in most of the triglyceride molecules of this group. One fraction (32.5 per cent. of the whole fat) consisted almost wholly of mono-saturated di-unsaturated glycerides, and palmitic acid formed over 80 per cent. of the saturated acids of this fraction. Hydrogenation at as low a temperature as possible gave a saturated product which on crystallisation yielded β -palmitodistearin (m.p. 67.7°C .; synthetic β -palmitodistearin has m.p. 68°C .). It is thus established that, in the monopalmito glycerides of cotton-seed oil, the palmitic group is exclusively attached to the β - or central hydroxyl group of the glyceryl radical.

D. G. H.

Determination of Iron and Copper in Butter. G. M. Motr and E. D. Andrews. (New Zealand J. Sci., 1940, 21, 249–265.)—

Iron.—Ten-g. samples of the butter are melted in 15-ml. centrifuge tubes, 1 ml. of 5 per cent. sodium hydrosulphite ($\text{Na}_2\text{S}_2\text{O}_4$) solution is added, and the tubes are shaken and left overnight at 35° – 40°C . One ml. of 20 per cent. trichloroacetic acid is introduced, the tubes are left for 30 minutes at 40° – 50°C ., and the melted fat is removed by suction. The aqueous layer is shaken with 5 drops of 10 per cent. sodium tungstate solution and filtered cold through papers washed, immediately before, with 5 per cent. nitric acid. The filtrate is shaken with 0.5 ml. of saturated potassium persulphate solution, and then with 2 drops each of nitric acid and hydrogen peroxide. One ml. of 30 per cent. ammonium thiocyanate is added, and the contents of all the tubes, including the standards, are made up to the same volume, and shaken with 2 ml. of amyl alcohol to extract the colour. To clear the amyl alcohol layer, the tubes are chilled in ice-water overnight, and the colours of the samples and standards are matched in a colorimeter. Very accurate results were obtained with the Klett bio-colorimeter. In 6 samples of butters the amounts of iron ranged from 0.3 to 0.9 p.p.m.

Copper.—Twenty-five-g. samples of the butter are melted at 40° – 50°C . in 50-ml. centrifuge tubes, and then each is treated with 5 drops of 3 per cent. hydrogen peroxide, 12 drops of conc. hydrochloric acid and 2.5 ml. of 20 per cent. trichloroacetic acid. The tubes

are well shaken and kept at 40°–50° C. for 30 minutes, after which the separated fat is removed. After cooling, 5 drops of 10 per cent. sodium tungstate are added to the aqueous layers in each of the tubes, and the tubes are again shaken and left for 30 minutes. Each sample is filtered, and the precipitate is washed with 3 ml. of 4 per cent. nitric acid and then with 5 per cent. trichloroacetic acid. The filtrate is treated with 2 ml. of 20 per cent. sodium citrate solution (to prevent precipitation of phosphate) and with a few drops of phenolphthalein solution. Sufficient ammonia (the same amount for each tube) to make the liquid alkaline is added, and each sample is made up to 40 ml. and treated with "a few grains" of solid sodium diethyl dithiocarbamate and 5 ml. of amyl alcohol. The tubes are shaken, allowed to stand for 3 or 4 hours, and again shaken, to complete the extraction of the coloured compound. The colours of the amyl alcohol extracts are then compared in a colorimeter with standards prepared from copper sulphate. In test experiments, in which known quantities of copper (0.1 to 0.5 p.p.m.) were added to a butter (copper content, 0.09 p.p.m.), the results were in close agreement with the calculated amounts. The copper-contents of 370 samples of creamery butter ranged from less than 0.1 to over 1.0 p.p.m. (1 sample); in 64 per cent. of the samples it did not exceed 0.2 p.p.m.

If the principal reagents are adequately purified, it is possible to obtain blank results so low that no appreciable error is caused by adding the reagents to the standards. For this purpose the trichloroacetic acid must be distilled *in vacuo*. The sodium tungstate and sodium citrate solutions are treated with a few mg. of sodium diethyl dithiocarbamate, followed by 2 or 3 extractions with amyl alcohol in a separating funnel. The aqueous solution is then ready for use.

Wet Oxidation Method.—More concordant results were obtained by the following modification of the wet oxidation method of W. Williams (*J. Dairy Res.*, 1931, 3, 93) than by the dry ashing procedure:—Twenty-five-gram samples of the butter are weighed into 50-ml. centrifuge tubes, 8 ml. of nitric acid (A.R. quality) are added to each, and the tubes are placed for about an hour in a boiling water-bath, their contents being stirred at intervals to ensure thorough mixing. The samples are cooled until bubbles of gas (nitrous fumes) cease to be evolved from the aqueous layer and are then centrifuged. Most of the fat is removed by means of a pipette attached to the water-pump, after which 5 ml. of petroleum spirit (b.p. 120° C.) are introduced, and this is removed in the same manner. The contents of each tube are transferred to a 200-ml. Kjeldahl flask, which is heated over a small flame until only about 1 ml. of liquid remains. Two ml. of sulphuric acid are introduced into each flask, and the heating is continued until nitrous fumes are no longer evolved; if necessary, the oxidation may be completed by adding a few drops of 30 per cent. hydrogen

peroxide. The crystalline residues are dissolved in water that has been redistilled from glass, and the solutions are transferred to Pyrex test-tubes, neutralised and treated with the reagents (citrate, etc.), and the analyses are completed in the colorimeter as before. Blank estimations are made simultaneously and a correction applied. Results thus obtained with 23 samples of butter (copper, 0.04 to 0.79 p.p.m.) agreed closely with those given by the filtration method.

Composition of the Fatty Acids in Chinese Coconut Oil. H. Nobori. (*J. Soc. Chem. Ind. Japan*, 1940, 43, 199–200).—The dried kernels of coconuts growing in Hainan Island, South China, contained 65.69 per cent. of oil with the following characteristics:—sp.gr. at 30/4° C., 0.9203; n_D^{20} , 1.4536; m.p., 24.2° C.; solidif. p., 22° C.; saponification value, 260.6; iodine value, Wijs, 8.7; Polenske value, 12.5; Reichert–Meissl value, 6.6; Hehner value, 90.7; thiocyanogen value, 6.7; acid value, 1.1; unsaponifiable matter, 0.38 per cent. The mixed fatty acids had sp.gr. at 30/4° C., 0.8999; m.p. 24.5° C.; solidif. p., 23° C.; n_D^{20} , 1.4410; saponification value, 268.4; iodine value, 9.9; neutralisation value, 263.5. The mixed fatty acids (500 g.) were esterified with an equal volume of methyl alcohol to which 2 per cent. sulphuric acid had been added and 492 g. of pale yellow liquid esters were recovered having sp.gr. at 20/4° C., 0.8776; n_D^{20} , 1.4352; saponification value, 254.1; iodine value, 9.5; acid value, 8.1. Twelve fractions were obtained by distillation of 470 g. of esters, and the residue was fractionated into a further three fractions. These fractions were refractionated in the usual manner, and the constants of the liberated fatty acids were determined. From the iodine and thiocyanogen values the fatty acids of the original coconut oil were calculated to contain: saturated acids, 92.2; oleic acid, 5.5; linolic acid, 2.3 per cent. Redistillation of the fatty acids from the higher-boiling fractions (14 and 15) indicated the presence of arachidic acid; each fatty acid fraction from 8 to 13 was separated into solid and liquid parts by the lead salt and alcohol method, but no hexadecenoic acid, $C_{16}H_{32}O_2$, found by Longenecker (*J. Biol. Chem.*, 1939, 130, 167), could be detected. The composition of the saturated fatty acids was calculated to be caprylic, 8.7; capric, 8.1; lauric, 51.3; myristic, 13.1; palmitic, 7.5; stearic, 2.0, with a small quantity of arachidic acid.

D. G. H.

Factors Affecting the Efficiency of Materials used in Packaging Frozen Foods. W. Rabak. (*Paper Trade J.*, 1940, 111, T.A.P.P.I. Sect., 110–112).—The resistance of carton boards to penetration by moisture may be measured from the increases in weight of anhydrous calcium chloride, which is placed in an air-tight container the lid of which is the carton material, in 6 days at

50 per cent. relative humidity and at 0° and 70° F. The calcium chloride (50 g.) was weighed into shallow weighing-bottles, which were placed in heavy-gauge aluminium containers (diameter 13.4 cm., depth 6.5 cm.), having a flat shoulder (width 1.8 cm.) on which were placed a pair of heavy rubber rings. The sample was then clamped tightly between the rings so as to seal the container. Results obtained at 0° and 70° F., respectively, were:—Coated boards (6 samples), 0.15, 0.78; impregnated boards (6 samples, types not specified), 0.24, 2.3 g. per 1000 sq. cm. per day. When a moist material was placed in direct contact with the carton (thickness, 0.020 in.) the figures obtained at 70° F. were:—Coated paperboard, 6.2; impregnated paper-board, 38.2; untreated cardboard, 110.2 g. of water per 1000 sq. cm. per day. Wax-treated papers were found to allow little penetration of moisture vapour, especially at low temperatures, whilst with vegetable parchment waxed on both sides the transmission was nil or negligible in 6 days. Creasing, however, produced marked increases in transmission, especially with waxed papers and at the higher temperatures. Moisture-proof viscose and laminated rubber materials were very efficient at all temperatures, even when creased. The losses in weight of loose frozen peas in various types of packages were determined by weighing at intervals for 9 months during storage at 0° and 15° F. under commercial conditions. The efficiency of the package in preventing losses in weight was found to depend mainly on the wrapping material used, but also on the efficiency of the seal. Satisfactory results were obtained with heavily-waxed paper, vapour-proof viscose and laminated rubber sheets, but ordinary viscose was less satisfactory. In general, a package which allows the loss of not more than 0.1 per cent. of the weight of the contents at 0° F. and 1.0 per cent. at 15° F. in 9 months, is regarded as satisfactory. J. G.

Biochemical

Human Foot Perspiration and Upper Boot Leather. A. Colin-Russ. (*J. Hyg. Camb.*, 1940, 40, 447; *J. Int. Soc. Leather Trades Chem.*, 1940, 24, 359.)—The effect of human perspiration on full chrome upper leathers tanned with basic chrome sulphate liquors is important in connection with the suitability of upper material for boots. The collagen complex of the leather is disrupted by the acidic and saline constituents of the perspiration, and solubilised chromium and ionisable sulphate are progressively released, the process being more pronounced at higher temperatures and when the tannage is more basic. The process may be regarded as double decomposition resulting in the formation of sodium sulphate, which may sometimes render the leather fibre brittle.

Estimation of Serum Iron and Pseudo-haemoglobin Iron with o-Phenanthroline. G. Barkan and B. S. Walker. (*J. Biol.*

Chem., 1940, 135, 37–42.)—Two ml. of blood or serum are measured into a short rimless test-tube, 1 ml. of 1.2 per cent. hydrochloric acid is added, and the tube is stoppered and kept at 37° C. for at least 1 hour. After cooling, 1 ml. of 20 per cent. trichloroacetic acid is added, and the mixture is allowed to stand for 1 hour at room temperature. The resulting suspension is centrifuged, and 2 ml. of the clear supernatant liquid are transferred to the cell of an Evelyn colorimeter. To the solution are added 0.5 ml. of saturated sodium acetate solution, 0.5 ml. of 1 per cent. hydrazine sulphate solution (to reduce the iron to the ferrous condition) in 2 M-acetate buffer solution (pH 4.5), and 0.5 ml. of 0.1 per cent. o-phenanthroline monohydrate solution. A blank solution, prepared by mixing 0.5 ml. of each of the above reagents with 0.5 ml. of the hydrochloric acid, 0.5 ml. of the trichloroacetic acid and 1 ml. of water, is introduced into another cell. The cells are stoppered and left for at least 1 hour at room temperature, after which 2.5 ml. of water are put into each cell, and the colours are measured in an Evelyn photoelectric colorimeter with filter 490. Ferric iron added to horse serum was estimated, with a maximum error of 4 per cent.

To determine pseudo-haemoglobin iron ("easily split off" blood iron), defibrinated blood, filtered through glass wool, is diluted with 4 volumes of water, and 10 ml. of this diluted blood are oxygenated by shaking in air, heated with 5 ml. of 1.2 per cent. hydrochloric acid, incubated at 37° C. for 16 to 24 hours and cooled, after which 5 ml. of 20 per cent. trichloroacetic acid are added, and the mixture is left for 1 hour at room temperature. It is then filtered and the filtrate is treated as described above to produce the red ferrous phenanthroline complex. F. A. R.

Improved Method for the Estimation of Non-haemin Iron. G. Brückmann and S. G. Zondek. (*J. Biol. Chem.*, 1940, 135, 23–30.)—One g. of the tissue pulp is ground in a mortar with powdered glass, 5 ml. of saturated sodium pyrophosphate solution and 10 ml. of 10 per cent. trichloroacetic acid. The mixture is transferred to a wide centrifuge tube, heated in a boiling water-bath for exactly 7 minutes and centrifuged. The residue is washed twice with 4 ml. of a mixture of the two reagents in equal parts. The combined centrifugate and washings are made up to volume, and aliquot portions are used for colorimetric estimations of iron either with thioglycolic acid or with o-phenanthroline. When great accuracy is required, the residue may be re-digested with sodium pyrophosphate solution and trichloroacetic acid, but the additional iron recovered in this way is seldom more than 5 per cent. of the total, and may usually be ignored. An extinction photometer (Leitz) with filter 495mμ is used for measuring the colours of the final solution. The results obtained by the method were compared with those obtained by digestion with pyrophosphate and trichloroacetic acid at room temperature for 3 days, and by the

hydrochloric acid extraction method of Starkenstein and Weden (*Arch. exp. Path. Pharmacol.*, 1928, 134, 274). In addition, the total iron-contents of the same tissues were measured after ashing with sulphuric and perchloric acids, and the haemin iron was estimated by the method of Yabusoe (*Biochem. Z.*, 1925, 157, 388): One g. of the tissue pulp is ground with powdered glass, 1 ml. of *N* hydrochloric acid and a few ml. of ice-cold absolute methyl alcohol. The mixture is centrifuged, and the residue is washed with methyl alcohol. The combined centrifugate and washings are shaken with 1 g. of powdered magnesium sulphate (iron-free), left for 30 minutes and centrifuged. The colour of the solution is compared with that of a haemin standard solution (filter 620 μ), or, better, its iron-content is estimated after ashing. The value so obtained may be subtracted from the total iron-content, giving yet another estimate of non-haemin iron content. The agreement between this value and that found by the new method was very close, the maximum divergence being 4 per cent. The hydrochloric acid method gave inconsistent results.

F. A. R.

Polariscopic Determination of Proteolytic Activity. Q. Landis. (*Cereal Chem.*, 1940, 17, 468-472; *J. Inst. Brewing*, 1940, 46, 380).—The method is based on the fact that the approximately linear decrease in the sp. rotation of gelatin as the temperature rises from 15° to 30° C., is affected by the action of proteolytic enzymes on the gelatin to an extent which depends on the enzymic activity. A fresh solution of gelatin is prepared by soaking 2 g. in 98 ml. of cold water and then warming the mixture to 40° C., an acetate buffer (to produce pH 4.8), and toluene (as preservative) being also added. A mixture of 25 ml. each of this solution and of an infusion of the enzyme in 2.5 per cent. sodium chloride solution (both at 40° C.) is digested in a 50-ml. stoppered Erlenmeyer flask for 5 hours at 40° C. If the solution is then turbid, it is cleared by immersion in boiling water until the soluble protein flocculates. The flask is then cooled rapidly, immersed in ice-water for 1 hour and then warmed to 20° C. and the value of $[\alpha]$ is determined; solutions which are turbid at this stage should be filtered. Blank solutions containing (a) no enzyme and (b) no gelatin should be made concurrently, but the former one should be poured into the polarimeter tube within 1 minute after cooling or it will gel. The following relationship between the concentration (c) of the enzyme (in milli-units per 50 ml. of reaction-mixture containing 25 ml. of the enzyme infusion) and the fall in rotation (p°) was deduced from experiments with a preparation of known proteolytic activity:

$$\log c = (0.728p + 0.378).$$

This holds for values of *c* ranging from 10 to 25. The original paper also contains a modified calibration method for use at high summer

temperatures, and tabulated values obtained with papain and with brewers' and distillers' malts.

J. G.

Determination of Tungsten in Biological Materials. J. C. Aull and F. W. Kinard. (*J. Biol. Chem.*, 1940, 135, 119-121).—The sample (e.g. 2 ml. of blood, 10 ml. of urine or 2 g. of organ) is heated gradually in a 500-ml. Kjeldahl flask with 4 ml. of conc. sulphuric acid, 10 ml. of a 1:1 mixture of conc. nitric acid and 70 to 72 per cent. perchloric acid, and a few glass beads. If charring occurs, more of the nitric and perchloric acid mixture is added until the solution clears. The heat is increased until white fumes are no longer evolved. After cooling, the solution is diluted with water and made distinctly alkaline to litmus with 40 per cent. sodium hydroxide solution. The solution is boiled vigorously for several minutes, cooled, transferred to a graduated flask of suitable size, and made up to volume. After filtering, a 5.0-ml. aliquot portion (containing not more than 0.1 mg. of tungsten) is measured into the tube of an Evelyn photoelectric colorimeter and the following reagents are added with shaking: 1.0 ml. of 15 per cent. potassium thiocyanate solution, 4.0 ml. of conc. hydrochloric acid and 1.0 ml. of titanium chloride reagent prepared by boiling 1.0 ml. of 10 per cent. titanous chloride solution with 2.0 ml. of conc. hydrochloric acid to expel traces of hydrogen sulphide and then diluting to 80 ml. with conc. hydrochloric acid. After 10 minutes the tubes are placed in the colorimeter and read against a blank tube containing reagents only, using filter 420. The tungsten-content is calculated by reference to a standard curve prepared from solutions containing 0 to 0.1 mg. of tungsten. The range of recoveries of added tungsten was from 95 to 105.8 per cent., with an average of 99.9 per cent.

F. A. R.

[Preparation of] Stable 2,6-Dichlorophenol-indophenol Solutions. I. Stone. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 415).—Solutions of sodium 2,6-dichlorophenol-indophenol used for the determination of vitamin C (*cf.* Mottern, Nelson and Walker, *ANALYST*, 1933, 58, 48) are relatively unstable, and must frequently be replenished and stored in the cold. Thus, after 8 days' storage of a solution at 2° and 30° C., the results of an assay fell, respectively, from 153 to 143 and from 153 to 90 mg. of reagent per 100 ml. of solution. Pre-extraction of the dry reagent with ether (*cf.* Knight, Dutcher and Guerrant, *Science*, 1939, 89, 184) did not effect much improvement. The following procedure is suggested:—Slightly more than the requisite weight of reagent (usually approximately 0.2 g. per 100 ml., for 0.005 *M* solutions) is stirred well with distilled dioxane (see below) containing 1 ml. of glacial acetic acid per 100 ml. After 15 minutes no more lumps should remain, and the solution is then filtered through a dry No. 42 Whatman paper. The clear red filtrate is standardised against pure ascorbic

acid in the usual way, the volume of dioxane added being less than 10 per cent. of the total volume in the titration-vessel, as larger proportions obscure the end-point. Since peroxides are produced in the commercial product if it is allowed to stand, 500 ml. of the dioxane should be distilled, the first and last 50 ml. being rejected. Traces of peroxides, however, may still remain, but they do not interfere with the vitamin C assay, although they affect the iodine and sulphuric acid method of standardisation (Menaker and Guerrant, *ANALYST*, 1938, 63, 201); hence the use of pure ascorbic acid for this purpose. The rate of evaporation of dioxane is not appreciably greater than that of water, and assays made at intervals during 3½ months with 0.005 *M* solutions (initial assay, 146 mg. per 100 ml.) showed decreases in strength of only about 2 mg. per month.

J. G.

Estimation of Aneurin by a Modification of the Melnick-Field Method. A. D. Emmett, G. Peacock and R. A. Brown. (*J. Biol. Chem.*, 1940, 135, 131-138.)—The method of estimating aneurin devised by Melnick and Field (*cf. ANALYST*, 1939, 64, 367) was applied to complex mixtures containing some of the other vitamins. The only modifications introduced were a simplified technique for adsorption and elution, and the substitution of the Lovibond tintometer for the colorimeter. Using pure solutions of aneurin, it was found that the concentration of the vitamin was directly proportional to the colour measured in red units and a standard curve was constructed from the data so obtained. The estimation of aneurin in tablets and ampoules gave results ranging from 95 to 103 per cent. of the theoretical value. The presence of other synthetic vitamins (riboflavin, vitamin B₆, nicotinic acid, pantothenic acid and ascorbic acid) had no effect on the accuracy of the aneurin assay, except when relatively much ascorbic acid was present, *e.g.* 2 mg. (aneurin 30γ). This interference by ascorbic acid could be overcome by cautious addition of either 0.01 *N* iodine solution or 1 per cent. potassium ferricyanide solution, but there is a risk that excess of the reagent will be added and this will attack the vitamin B₆. A better method of overcoming the interference is to adsorb the aneurin on "Superfiltrol" (see below). The following pharmaceutical preparations were assayed by the direct method: wheat germ extract, emulsion of vitamins A, B and D, fortified liver extract, stomach extract with vitamin B₆. With the exception of the liver extract, the results obtained were in close agreement with the values obtained by biological assay. In some instances the vitamin may be present either in a complex soluble mixture or in a combined form, as in yeast, and it is necessary to digest the material to split off the vitamin; interfering factors must then be removed and this is best done by adsorption of the aneurin. A stock solution of the test material is prepared, of such a concentration that it contains between 5 and 20γ

of aneurin per ml. Of this solution, 2-, 3- and 4-ml. portions are transferred to three 15-ml. centrifuge tubes and diluted to 5 ml. The pH is adjusted to 4-5 with dilute hydrochloric acid, 0.1 to 0.15 g. of "Superfiltrol" is added, and the tubes are shaken intermittently for an hour; they are then centrifuged and the supernatant liquid is discarded. To each of the adsorbates are added 3 ml. of water and 3 ml. of 95 per cent. alcohol containing 5 mg. of phenol per ml. The pH is then adjusted to 7-8 (thymol blue) with dilute sodium hydroxide solution, and 6 ml. of the diazo reagent are added. The mixture is left for at least 2 hours at room temperature and then filtered on a small Hirsch filter. The adsorbate is washed with 5 ml. of water and transferred, with the filter-paper, to a dry centrifuge tube, and 2 ml. of 95 per cent. alcohol are added. The tube is stoppered, shaken to elute the pigment, and then centrifuged. The supernatant liquid is transferred to a tintometer cell, and the colour is measured, the results being calculated from the standard curve. Whereas low results were obtained with a dried yeast, a wheat germ extract and an elixir by the direct assay method, results given by the adsorption method agreed closely with the biological values for these products.

F. A. R.

Chemical Estimation of Nicotinic Acid: Inhibitory Effect of Cyanogen Bromide upon the Aniline Side Reactions. D. Melnick and H. Field. (*J. Biol. Chem.*, 1940, 135, 53-58.)—In the method previously described (*cf. ANALYST*, 1939, 64, 367) for the estimation of nicotinic acid in biological material by reaction with cyanogen bromide and aniline, a blank was prepared by simply diluting the test solution with an alcoholic buffer solution. Harris and Raymond (*Biochem. J.*, 1939, 33, 2037), however, advocated the addition of the base (in their method *p*-aminocetophenone) to the blank solution, as it was found that the base reacted with substances in the hydrolysates to produce colours indistinguishable from that obtained with nicotinic acid. It has now been found that aniline behaves in a similar manner, but that in presence of cyanogen bromide the interfering side-reaction does not occur. Thus the addition of aniline to the blank solution is not recommended.

F. A. R.

Assay Method for Pantothenic Acid. D. Pennington, E. E. Snell and R. J. Williams. (*J. Biol. Chem.*, 1940, 135, 213-222.)—The pantothenic acid content of tissue extracts can be estimated by measuring the growth-response of a culture of *Lactobacillus casei* *s. s.* A basal medium containing alkali-treated peptone, glucose, sodium acetate, alkali-treated yeast extract, acid-hydrolysed casein, cystine, riboflavin and inorganic salts is prepared and the pH is adjusted to 6.6 to 6.8; 5-ml. portions of the medium are introduced into each of 10 bacteriological test-tubes. To some of the tubes are added volumes up to

5 ml. of a standard pantothenic acid solution and to others the samples for analysis. The tubes are sterilised, cooled and inoculated with 1 drop of a 24-hour culture of the organism in normal saline. The tubes are then incubated at 37° C. for 72 hours. The response of the organisms to pantothenic acid can be measured either by titrating the lactic acid produced during growth or by measuring the turbidity in a thermoelectric turbidimeter or in an Evelyn photoelectric colorimeter. The response, by both methods of measurement, was found to be nearly directly proportional to the amount of pantothenic acid present when this was between 0.05 and 1.2 mg. units (1 mg. unit = 0.08% of calcium *d*(+)-pantothenate). A standard curve must be prepared with each set of assays. With pure solutions of pantothenic acid, recoveries of 105 to 109 per cent. were obtained by the titrimetric method and of 94 to 105 per cent. by the turbidimetric method. Recoveries of a similar order were obtained when pantothenic acid was added to yeast extract, liver extract or whey; such extracts were prepared by autoclaving, preferably after autolysis under benzene. Some substances, however, did not show such good recoveries and the assay figures did not agree when calculated from different doses. The most important of these substances was urine; others were oysters and mushrooms. The error could be rectified by adding to the basal medium a supplement prepared by autoclaving the sample in question with sulphuric acid, and then removing the sulphuric acid by treatment with baryta. F. A. R.

Antiascorbic Values of Fruits and Vegetables. M. Olliver. (*Lancet*, 1940, II, 190-195).—The following values were found for the amounts of ascorbic acid (mg. per 100 g.) in a number of fruits and vegetables as normally consumed. (a) Raw fruits and vegetables: watercress, 58 to 72; strawberry, 44 to 93; orange-juice, 33 to 77; lemon-juice, 29 to 60; grapefruit-juice, 28 to 64; raspberry, 21 to 37; tomato, 12 to 42; lettuce, 5 to 30; apple, 4 to 17; banana, 6 to 8; melon, 2 to 3; pear, 2 to 3. (b) Home-cooked or commercially canned fruit and vegetables: black currant, 80 to 152; brussels sprouts, 19 to 44; asparagus 13 to 44; strawberry, 19 to 35; spinach, 9 to 50; cauliflower, 16 to 42; gooseberry, 13 to 31; cabbage, 11 to 33; potato (new), 10 to 20; turnip, 11 to 17; pea, 6 to 20; raspberry, 4 to 17; potato (old), 7 to 10; carrot, 1 to 15; stringless bean, 2 to 6; greengage, apple, plum, 1 to 4; apricot (dried), 1 to 2; prune (dried), 1. With the exception of potatoes, the outer tissues of fruits and vegetables generally contain more ascorbic acid than the inner tissues. Ordinary methods of drying fruits readily destroy vitamin C, whereas cooking or canning cause only a partial loss. In household cooking this loss is brought about not so much by the effect of temperature as by extraction of the vitamin in the liquor; with fruit, the syrup or water is consumed, but with vegetables the liquor is thrown away. The more water

used in boiling the vegetables and the longer the time of heating, the greater is the loss of vitamin. The presence of alkali, however, does not increase this loss, contrary to earlier work which suggested that the addition of soda to boiling greens had an adverse effect on the ascorbic acid. In canning there is little loss of ascorbic acid during the cooking process, most of the loss observed occurring during the preliminary blanching or hot-water treatment used to destroy enzymes; the loss by extraction is less than in home-cooking because less water is used. The material used by the canner must be fresh, whereas that purchased by the housewife has usually been stored for some time, and it has now been shown that considerable losses of ascorbic acid occur during storage. Thus, canned fruits and vegetables tend to contain more of the vitamin than the corresponding materials purchased in shops and cooked at home. Vitamin C, contrary to certain beliefs, withstands the heat treatment of jam boiling, the apparent loss observed being due chiefly to dilution with added sugar and water. The costs of vitamin C from various sources are compared. F. A. R.

Methylene Blue Reducing System of Palestine Orange Peels Investigated by the Thunberg System. L. Frankenthal. (*Enzymologia*, 1939, 6, 287-306).—Peel juice, prepared with the aid of a hydraulic press, was decolorised with animal charcoal. The filtrate had a high reducing activity towards methylene blue in presence of phosphate; this could not be due to ascorbic acid, which is adsorbed by animal charcoal. In absence of phosphate, reduction was very slow. The reducing substance was heat-labile, unlike that present in orange-juice, but, notwithstanding its heat-lability, it passed readily through membranes, thus lacking the colloidal nature ascribed to enzymes. The properties of the substance were compared with those of a number of reducing systems, and were found to agree very closely with those of dehydroascorbic acid and its more stable isomer, 2:3-diketogulonic acid. The presence of dehydroascorbic acid in orange peel juice was confirmed (a) by incubating the juice with glutathione (this produced ascorbic acid, which was then titrated with dichlorophenolindophenol) and (b) by isolating the 2:4-dinitrophenyllosazone of dehydroascorbic acid, m.p. 271 to 273° C. It was also shown that the dehydroascorbic acid was actually present as such in the peel and not produced subsequently either by catalytic oxidation by traces of metal or as a result of the charcoal treatment. Dehydroascorbic acid was found not to be stable in the juice for long, but changed into diketogulonic acid, this being responsible for the reduction of the methylene blue; old juice contained no dehydroascorbic acid. No dehydroascorbic acid was found in orange juice. The results suggest that orange peel contains an enzyme capable of oxidising ascorbic acid to dehydroascorbic acid. F. A. R.

Erratum. September issue, p. 519 (Estimation of Phosphorus), line 12 of the Abstract. For "about 0.4 mg. of phosphorus" read "not more than 0.4 mg. of phosphorus."

Organic

Determination of Hydroxyl Groups with the Grignard Reagent. W. Fuchs, N. H. Ishler and A. G. Sandhoff. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 507-509.)—The method of Zerewitinoff (*Ber.*, 1907, 40, 2023; 1908, 41, 2236) for the determination of active hydrogen based upon the evolution of methane in reactions with magnesium methyl iodide is not highly accurate, and the improved method of Kohler, Stone and Fuson (*J. Amer. Chem. Soc.*, 1927, 49, 3181) is too intricate for occasional use. It was found necessary therefore to devise a simpler and more rapid method based upon that of Zerewitinoff. The apparatus consists of a reaction vessel connected with a manometer and a gas burette. The reaction vessel consists of a lower portion containing the reagent and an upper portion carrying a vertical capillary tube and two horizontal side tubes connected with manometer and gas burette in the usual manner. The upper portion fits over the lower portion by means of a ground-glass joint and is firmly held in position by brass springs. The apparatus differs from conventional design chiefly in that the sample is weighed into steel cups, which are suspended from protuberances on the vertical capillary tube in such a manner that they can be lowered into the reagent by means of an electromagnet. The air and moisture in the apparatus are replaced by pure dry nitrogen. No volume changes occur during manipulation, and a dibutyl phthalate manometer makes pressure adjustments rapid and accurate. The sample, which has been dried at 100° C. in a 2-mm. vacuum (or if volatile at a lower temperature and higher pressure), is weighed into two steel cups which are then hung on the capillary tube in the reaction vessel containing the Grignard reagent dissolved in 15 to 20 ml. of iso-amyl ether. The apparatus is quickly fitted together and flushed out with dry nitrogen which has been passed through iso-amyl ether. The reaction vessel is then surrounded with a water-bath at 70° C., the flushing with nitrogen is continued and the gas burette is filled with nitrogen. The water-bath at 70° C. is replaced by one at room temperature, and the system is adjusted repeatedly to atmospheric pressure until pressure conditions remain stable for 5 or 10 minutes. One of the cups is then lowered into the reagent, and the reaction, which is usually vigorous, is allowed to proceed. After reaction has ceased the usual procedure in gas analysis is followed, and the methane evolved is measured. The second portion of sample serves for a duplicate determination, and its cup can be lowered into the reagent as soon as the first determination is completed. Calculations were based on the density of methane at the

temperature at which the system was maintained during the determination. Grignard reagent prepared with *n*-butyl ether as solvent reacted more quickly than the reagent in iso-amyl ether, with equally precise results, but pyridine was found unsuitable as a solvent. Benzoic acid, anisic acid, α -naphthol and β -naphthol gave results closely in accord with theory. The results obtained with acetoacetic ester and malonic ester agreed with the theoretical values for the completely enolised compounds. The toluic acids gave high results, probably due to impurities. The three phthalic acids, trimesic acid, pyromellitic acid and benzene pentacarboxylic acid did not yield methane with the reagent, the apparent explanation being that, since these compounds are insoluble in the medium, the Grignard reagent does not react in heterogeneous systems. Picric acid did not react, probably owing to its insolubility, but tribromophenol reacted completely. Resorcinol, hydroquinone, toluhydroquinone and phloroglucinol either failed to react or reacted incompletely. Their solubility was slight and they are known to be capable of keto-enol tautomerism. Resorcinol in *n*-butyl ether solution apparently reacts as a monoketomonoenol compound. A. O. J.

Photometric Estimation of Furfural. R. A. Stillings and B. L. Browning. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 499-502.)—The colorimetric method for the estimation of furfural by means of the red colour produced with salts of aniline or xylidene has the advantage over the gravimetric and volumetric methods that the methylfurfural and hydroxymethylfurfural, always present in the distillates from plant tissues, are not included in the determination. For transmission measurements a General Electric recording spectrophotometer was used, and for photometric measurements an absorption meter and a Weston Photronic cell connected through a potentiometer with a microammeter. A Jena BG 18 glass filter was fitted to the opening of the photronic cell. Glass cells providing a thickness of liquid of 5 mm. were used. Owing to the difficulty of obtaining pure xylidene or its isomers (or constant mixtures of these) the reaction with aniline acetate was considered the more satisfactory for photometric work. A study of the effects of variations of temperature and of the concentrations of acetic acid, aniline, sodium chloride and furfural yielded the following results:—Increase of acetic acid concentration considerably increased the stability of the colour, and a concentration of 200 to 500 g. per litre proved satisfactory. Neutralisation of the distillate prepared by the usual methods (containing about 12 per cent. of hydrochloric acid) introduced up to 200 g. of sodium chloride per litre, and the final concentration after the addition of other reagents was 20 g. per litre. It is recommended that this concentration should be maintained. The statement of other investigators (Barta, *Biochem.*

Z., 1934, 274, 212; Riffart and Keller, *Z. Unters. Lebensm.*, 1934, 68, 113; Tolman and Trescott, *J. Amer. Chem. Soc.*, 1906, 28, 1619; Abst., ANALYST, 1907, 32, 49) that Beer's Law is valid for the furfural-aniline colour was confirmed for furfural concentrations of 0.5 to 4.5 mg. per litre in presence of the above-mentioned amount of sodium chloride, and under these conditions a concentration of aniline of 50 g. per litre provided high sensitivity, reasonable stability and relatively slight variations in colour with variations in aniline concentration. Rise of temperature decreased the stability of the colour and the time required to reach the minimum transmission. The transmission at its minimum value is not affected over the range 15° to 30° C., but for precise work the temperature should be controlled, preferably at 20° ± 0.5° C., at which temperature minimum transmission is reached in 55 minutes. The error introduced by presence of methylfurfural and hydroxymethylfurfural is less than 1 per cent. if the concentration of these substances is less than that of the furfural, as it is in the usual pentosan distillates. The method is as follows:—The solution containing 0.05 to 0.45 mg. of furfural is exactly neutralised to phenolphthalein with 10 per cent. sodium hydroxide solution and, after dilution of the liquid with water to about 40 ml., sufficient sodium chloride solution (200 g. per litre) is added to produce a final concentration of 20 g. per litre. Freshly distilled aniline (5 g.) is added by means of a specially graduated pipette to 50 ml. of glacial acetic acid and, after adjustment of the temperature to 20° C., this reagent is added to the liquid, which is then diluted with water to 100 ml. and stored in the dark at 20° ± 0.5° C. for 55 minutes. The transmission is then measured, and a control determination is made with the reagents by the same procedure; the result of this is taken to be equivalent to 100 per cent. of furfural. The quantity of furfural present in the sample is found by means of a calibration curve constructed for the instrument.

A. O. J.

Linolenic Acid in the Leaves of Land Plants. M. Tsujimoto. (*J. Soc. Chem. Ind. Japan*, 1940, 43, 208-209.)—The leaves of certain land plants were dried and extracted with ether or petroleum spirit, the extract was saponified with alcoholic potassium hydroxide, the unsaponifiable matter was removed, and the fatty acids obtained by decomposition with hydrochloric acid were dissolved in ether. The solution was chilled and treated with a very large excess of bromine, and the precipitated bromides were collected, weighed and analysed. The bromide yields (as per cent. of the ethereal extracts) were as follows:—*Sasa paniculata* (Nemagardake), 2.6; *Sasa albo-marginata* (Kumazasa), —; *Pinus Thunbergii* (black pine), 0; *Ginkgo biloba*, green leaves, 0.5; yellow leaves, 0.7; *Platanus orientalis*, 1.6; *Trifolium repens* (clover), 2.0. The linolenic bromide from the

leaf oil of *Sasa paniculata* melted at 177° to 178° C. and contained 63.28 per cent. of bromine. Except in the oil from *Pinus Thunbergii*, linolenic acid was present in all the leaf oils, but as far as the present experiments go, highly unsaturated acids, similar to those present in fish oils and in algae (*cf. Chem. Umschau*, 1925, 32, 125), are not present.

D. G. H.

Determination of Sludge in Used or Oxidised Motor Oils. C., G. Ludeman. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 520-525.)—Motor oil sludge is composed of the alteration products of the oil, dust, dirt, metal wearings, emulsions, etc., but this investigation was confined to the alteration products formed by oxidation and polymerisation, and the oils used were artificially sludged by blowing air through them at 175° C. The components of the sludge may be grouped into asphaltenes (bitumens soluble in carbon disulphide but insoluble in paraffin naphtha), carbenes (bitumens soluble in carbon disulphide but insoluble in carbon tetrachloride) and carboids (insoluble in carbon disulphide). About 10 ml. of oil were weighed in an Erlenmeyer flask, and the sludge was precipitated by adding 200 ml. of solvent a portion of which was retained for washing down the sides of the flask. The tightly stoppered flask was set aside in the dark at room temperature (about 25° C.) and, after exactly 24 hours the contents were filtered through a 30-ml. Coors No. 4 Gooch crucible which contained a mat, 3 mm. thick, of Powminco asbestos and had been thoroughly dried at 110° C. The supernatant liquid was passed through the crucible first, the sludge being retained as far as possible and added all together at the end. When the contents of the flask had been completely transferred, the cake of sludge was sucked dry until it was broken by cracks. During the washing of the crucible with the solvent the suction was interrupted three times for two minutes while the crucible was half-full of solvent. Usually 150 to 200 ml. of solvent were required for washing. After the final washing the crucible was dried at 110° C. for an hour and weighed. A number of solvents were examined for their precipitating properties, and isopentane was found to precipitate the greatest amount of sludge. An attempt to relate the chemical and physical properties of solvents with their precipitating power was unsuccessful, and, since the possibility of the exact reproduction of solvents consisting of the various naphthas from crude sources is small, it is recommended that definite chemical compounds, such as isopentane, carbon tetrachloride and carbon disulphide, should be used as standard precipitants. A fourth reproducible solvent is propane, as recommended by Hall, Levin and McMillan (*Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 183). The following is a complete analytical procedure for used or oxidised motor oils:—(a) asphaltenes, carbenes and carboids with inorganic material by precipitation with isopentane; (b) carbenes and carboids with

inorganic matter by precipitation with carbon tetrachloride; (c) carboids with inorganic matter by precipitation with carbon disulphide; (d) soluble sludge by propane precipitation of the oil obtained by evaporation (preferably *in vacuo*) of the filtrate from (a); (e) dissolved sludge by propane precipitation of an oil sample filtered at 25° C., with or without pressure; (f) inorganic material by ignition of the precipitates (a), (b) or (c). The individual components are then found as follows:—Soluble sludge is given directly by (d); asphaltenes are given by the difference between (a) and (b); carbonies by the difference between (b) and (c); carboids by the difference between (c) and (f); inorganic material is given directly by (f); undissolved sludge is given by the difference between (a) and (e), and dissolved sludge directly by (e). The sludge was also determined by a volumetric method based upon its separation by precipitation and centrifuging by a standardised procedure, but there appeared to be no simple relation between the sludge found by this method and that found by the gravimetric method.

A. O. J.

Effect of Glycerin on the Distillation Method for Water. R. B. Trusler. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 509–510.)—

Xylene is frequently recommended as the distillation medium in the determination of water in soap by the method of Dean and Stark (*J. Ind. Eng. Chem.*, 1920, 12, 486; *Abst.*, *ANALYST*, 1920, 45, 270), but it is necessary to determine whether soaps containing glycerin (e.g. liquid potassium soap, potassium-vegetable oil soap and the so-called cold-made soaps) behave in the same manner towards the distillation medium as do the majority of bar, flaked and powdered soaps containing no glycerin. In order to determine the effect of the presence of glycerin, water was added to weighed quantities of anhydrous glycerol and the distillation was carried out with the precautions described in a previous paper (*Oil and Soap*, 1939, 16, 239). The data obtained with benzene were similar to those obtained with toluene, the maximum average error obtained when 4 to 10 ml. of water were added to 10 g. portions of glycerol being +0.095 ml. with benzene and +0.17 with toluene. With these media the error increases with increase of water. With xylene the absolute error is greatest in absence of water, and least when the weights of water and glycerol are approximately equal. Since the b.p. of xylene is much higher than that of benzene or toluene some glycerin is undoubtedly carried over with the xylene vapour. The difference between water determinations made with benzene and those made with toluene did not exceed 0.1 per cent. and, since dehydration is more rapid with toluene, analytical comparisons were made only between toluene and xylene as distillation media. A series of moisture determinations made upon a number of soaps showed that with xylene the error per cent. of sample ranged from 0.05 for an "all soda" powdered laundry soap to

1.10 for a cold made coco-olive bar soap. When xylene is used instead of toluene, therefore, the error in the amount of water found may exceed 1 per cent. A greater error will be observed in the analysis of soaps made with fatty oils of the coconut oil type than with soaps made from longer chain fatty acids since the former type yield more glycerol. Toluene and xylene gave identical results with soaps free from glycerin. In order to obtain universal agreement in the determination of anhydrous soap by the distillation method, toluene should be used as the distillation medium, and the commercial variety known as 10° toluene is recommended. A. O. J.

Detection and Determination of Ethylene Glycol in Lubricating Oil. H. Lamprey, E. E. Sommer and A. D. Kiffer. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 526–527.)—

The examination of crank case oil for anti-freezing materials is sometimes necessary to detect leakage of engine-cooling solution into the crank case and to determine the effect of such substances upon sludge formation in the crank case. The most common anti freezing materials are methanol, ethanol and ethylene glycol, although glycerin, diethylene glycol, propylene glycol, isopropanol, sugars and inorganic salts are sometimes used. Specific methods for the identification of methanol and ethanol are known (Levin, Uhrig and Stehr, *Ind. Eng. Chem., Anal. Ed.*, 1939, 11, 134), but no satisfactory procedure for the detection or determination of ethylene glycol has been described. The method now suggested is based upon isolation of the glycol and its identification by its physical properties. To 100 ml. of the oil, 100 ml. of xylene are added, and the mixture is distilled for 2 hours by the method described in "*Standards in Petroleum Products and Lubricants*," p. 345 (American Society for Testing Materials, Philadelphia, 1937). If calcium chloride or oxalic acid has been detected, 1 g. of sodium carbonate should be added to the mixture before distillation. The lower aqueous layer of the distillate is fractionated in a small column to separate the ethylene glycol, a micro equipment being used if the concentration of the glycol is low (Morton, "*Laboratory Technique in Organic Chemistry*," 1st Ed., pp. 50, 73, 74; New York, McGraw Hill Book Co., 1938). In the rare event of the occurrence of more than one anti-freezing agent, further fractionation will be necessary. The ethylene glycol is finally identified by determination of the refractive index and sp. gr. of the fraction boiling within the range 195° to 200° C., and tables for this purpose are given in the original paper. If no other high boiling anti-freezing agent is present in the distillate, the method may be made quantitative by measuring the volume and n_D (or volume and sp. gr.) of the aqueous portion of the distillate. The following anti-freezing agents do not interfere with the method:—calcium chloride, oxalic acid, glycerin, sugar and diethylene glycol. Later experiments have shown that ethylene glycol

may be identified in presence of much larger quantities of propylene glycol, diethylene glycol and methyl and ethyl cellosolves. The sensitivity of the method may be increased by increasing the amount of sample originally taken.

A. O. J.

Identification of Aromatic Halogenonitro Compounds as Piperidyl Derivatives. M. K. Seikel. (*J. Amer. Chem. Soc.*, 1940, **62**, 750-753.)—Piperidine reacts more readily than other bases, such as ammonia, aromatic amines or morpholine, with compounds containing reactive halogen derivatives, while the reaction products crystallise readily and, in general, contain few by-products. Two general procedures have been developed: (A) in which the compound is heated for an hour under reflux with piperidine, and (B) in which an alcoholic solution is heated with piperidine for 15 minutes on the steam-bath. Procedure A yielded identifiable compounds in 79 per cent. of the 37 halogen nitro compounds studied, and procedure B in 74 per cent. Definite piperidyl derivatives were obtained with a large number of the compounds. In procedure A, 0.5 g. of the halogenonitro compound is treated with 1.5 ml. (1.0 ml. for bromo compounds) of piperidine, and the mixture is heated under reflux in an oil-bath for 1 hour, cooled, treated with water and filtered. If the product is an oil, it may be made to crystallise by chilling with ice. The paper gives the melting-points and other characteristic properties of the possible piperidyl derivatives, together with observations which will be of assistance in identifying the aromatic halogenonitro compounds studied.

Detection of Aromatics in Air. G. R. Gilbert and R. E. Tannich. (*Ind. Eng. Chem., Anal. Ed.*, 1940, **12**, 433-436.)—Existing methods (listed by Yant, Pearce and Schrenk, *U.S. Bur. Mines, Rept. Investigations*, 1936, 3323) are unsuitable for small proportions of aromatic hydrocarbons, or necessitate the use of an interferometer or spectrograph, and refractometric methods involve the use of low temperatures, with the result that ice from atmospheric moisture blocks the apparatus. In the method suggested, blends of known volumes of methyl alcohol vapour with known volumes (*a*) of the sample and (*b*) of air containing known volumes of the aromatic hydrocarbons, are condensed by cooling, and the *n* of each condensate is determined in an Abbé refractometer at identical temperatures. Methyl alcohol is used because its *n* (while less than that of the aromatic hydrocarbons) is similar to that of water, and any errors due to variations in the humidity of the sample of air are thereby minimised. Several samples of the air are obtained in 5-litre, short-necked Pyrex flasks fitted with a rubber stopper containing a thermometer and 2 tubes with stop-cocks (diameter, 4 mm.), which reach just below the neck. If the flasks are evacuated (to about 2 mm. of mercury, abs. pressure) the sample may be taken by inserting the outer end of one of the tubes in

the air under investigation and opening the stop-cock. The temperature is then recorded. Exactly 0.5 ml. of methyl alcohol is introduced into the flask through one of the tubes and evaporated by warming the closed flask to approximately 38° C.; the contents of the flask are then condensed by cooling to -56.7° C., and the *n* of the condensate (which is removed in a long fine capillary pipette) is determined. Solutions containing 0.1, 0.5, 1.25, and 2.5 ml. of the liquid contaminant in 100 ml. of methyl alcohol are next prepared, and 0.5 ml. of each is measured into an evacuated flask, the vacuum being then broken by admission of air (free from hydrocarbons). This procedure (heating and cooling) is repeated, and the values of *n* obtained are plotted against the corresponding volumes of contaminant originally present in the 0.5 ml. of solution, so as to produce a calibration curve. A portion of the liquid responsible for the contamination is distilled (Engler), and its aromatic content and density at 15.6° C. before and after extraction with 4 volumes of 99.5 per cent. sulphuric acid (*d* and *d'*, respectively) are determined. Then *d'*, the density (at 15.6° C.) of aromatic hydrocarbons present in the liquid contaminant = $[d - (1 - x)d']/x$, where *x* is the aromatic content of the liquid sample (expressed as a fraction, in terms of vol.). Also the aromatic content (in p.p.m. of vapour by vol.) is given by $9120cd'(460 + t)760/pm$, where *c* is the volume of total hydrocarbons (expressed in ml. of liquid) present in the sample of air (as read from the calibration curve); *m* the mol. wt. of the aromatic hydrocarbons; *t* the temperature (in °F.) and *p* the abs. pressure (in mm. of mercury) of the contaminated air. The derivation of the above equations and worked examples are set out in the original paper. For concentrations of aromatic hydrocarbons of the orders of 100 and 400 p.p.m., the errors are approximately 15 and 80 p.p.m., respectively.

J. G.

Separation and Determination of 2,4-Diaminodiphenylamine. I. S. Shupe. (*J. Assoc. Off. Agr. Chem.*, 1940, **23**, 719-721.)—This constituent of certain hair dyes is chemically similar to *p*- and *m*-phenylenediamines. The free base is said to be more stable in air than its salts, but aqueous solution of base and salts are rapidly oxidised to form brown to purple solutions. When purified by sublimation *in vacuo* it has m.p. 130° to 131° C. The method for its determination is based upon its solubility in carbon tetrachloride, in which most of the diamines are insoluble. The aqueous solution (25 ml.) is made alkaline with excess of sodium bicarbonate, 0.1 g. of sodium sulphite is added to inhibit oxidation, and the liquid is extracted immediately with five 20-ml. portions of carbon tetrachloride. The extracts are washed with two 20-ml. portions of water, and the washing water is extracted with another portion of the solvent. The combined carbon tetrachloride extracts are filtered through cotton-wool, evaporated on a steam-bath and dried in a desiccator.

The residue may be dried at 100° C. for 30 minutes without appreciable decomposition, but this yields a discoloured product. If aminophenols are present, the carbon tetrachloride extract is given a preliminary washing with 25 per cent. sodium hydroxide solution. It is important that extraction with carbon tetrachloride should be made immediately after rendering the solution alkaline, because the base rapidly becomes crystalline and difficult to extract. Although the diamino-diphenylamine is almost insoluble in petroleum spirit whereas 4-aminophenylamine is soluble, attempts to separate these compounds by means of this solvent were not completely successful, apparently because the diamino-diphenylamine crystallises out and carries with it some of the 4-aminodiphenylamine. The residual crystalline material, dissolved in hydrochloric acid and reprecipitated with sodium bicarbonate gave an additional yield of the 4-amino compound when extracted again with petroleum spirit. Carbon tetrachloride extracts both of these compounds from alkaline solutions.

A. O. J.

Determination of Thioglycollates in Depilatories. E. M. Hoshall. (*J. Assoc. Off. Agr. Chem.*, 1940, 23, 727-734).—Of the organic sulphur compounds (including calcium thioglycollate, calcium dithioglycollate, calcium thiolactate, the calcium salt of pseudo-thiohydantoin and the aliphatic mercaptans) that occur in modern depilatory preparations, calcium thioglycollate appears to be used most frequently. The following method serves for its determination:—If the sample is a non-oily paste, 3 to 5 g. are washed with warm water through a filter into an iodine flask. If the sample is an oil-in-water emulsion, a portion (4 to 10 g.) is shaken with 100 ml. of water until no lumps remain, and the mixture is diluted to 200 ml. and filtered, the first 20 ml. of the filtrate being rejected. The filtrate (100 ml.) is then transferred to an iodine flask. Depilatories rarely occur as water-in-oil emulsions, but probably the oil phase could be removed by extraction with chloroform and the residue treated by either of the above-mentioned methods. The clear liquid is slightly acidified to litmus with 0.1 *N* hydrochloric acid, the flask is quickly closed and shaken, and the stopper is sealed with 0.1 *N* iodine solution. The stopper is gently opened, the iodine is allowed to run in and titration is continued with 0.1 *N* iodine solution in presence of starch. Each ml. of iodine solution is equivalent to 0.018423 g. of calcium thioglycollate with 3 molecules of water of crystallisation. Satisfactory results were obtained by this method with depilatory pastes containing 4 to 6 per cent. of calcium thioglycollate made in imitation of commercial preparations. Calcium thioglycollate reacts quantitatively with one-half molecule of iodine in neutral solution. The reaction has not been investigated, but it appears to be similar to the oxidation of cysteine to cystine. In order to detect calcium thioglycollate in depilatories,

the following tests may be applied to the clear filtrate obtained from an aqueous suspension:—With 5 per cent. copper sulphate solution a violet-brown precipitate, soluble in dilute acids and in ammonia, is formed. With a few drops of 0.1 *N* ferric chloride solution, an evanescent indigo blue colour appears in neutral or slightly acid solutions, and on further addition of the reagent the colour changes to green, then to purple-brown and finally to yellow with formation of a precipitate. The test used by Hamence (*ANALYST*, 1940, 65, 152) to distinguish molybdenum from tungsten with thioglycollic acid may be adapted to the detection of thioglycollates. The dilute solution of the thioglycollate is treated with 1 ml. of 3 per cent. ammonium molybdate solution and 0.5 ml. of hydrochloric acid, and the mixture is diluted to 50 ml. A greenish yellow to greenish yellow-brown colour forms and becomes green, with separation of a precipitate, on addition of ammonia. The usual components of thioglycollate depilatory preparations do not interfere with these tests.

A. O. J.

Separation of *p*-Aminophenol, *p*-Methylaminophenol and *o*-Aminophenol. I. S. Shupe. (*J. Assoc. Off. Agr. Chem.*, 1940, 23, 721-727).—These aminophenols are commonly used in certain classes of hair dyes. To determine *p*-methylaminophenol in presence of *p*-aminophenol and *o*-aminophenol a portion not exceeding 400 mg. of the mixture is treated with 5 ml. of 10 per cent. hydrochloric acid and sufficient water to make 15 ml. The mixture is cooled to 15° C., mixed with 5 ml. of 10 per cent. sodium nitrite solution and extracted with five 20-ml. portions of ether free from alcohol and peroxides. The first four extracts are washed with 15 ml. of water, then with 10 ml. of saturated sodium bicarbonate solution and finally with 15 ml. of water, and the three washing solutions are shaken individually with the fifth ethereal extract. The combined ethereal extracts are filtered through cotton-wool, the greater part of the solvent is removed on the water-bath, and the last portion is allowed to evaporate spontaneously. The residual *p*-nitrosomethylaminophenol is dried in a desiccator and weighed, and the weight is multiplied by 0.809 to obtain the corresponding weight of *p*-methylaminophenol. To determine *p*-aminophenol in presence of *o*-aminophenol and *p*-methylaminophenol, the mixture (not exceeding 200 mg.) is treated in a stoppered vessel with 5 ml. of 10 per cent. sodium acetate solution, 50 mg. of powdered sodium bisulphite, and finally with 0.5 ml. of benzaldehyde for each 50 mg. of sample. The mixture is heated for 5 minutes on the steam-bath and, when cold, is shaken thoroughly with 25 ml. of petroleum spirit and allowed to stand at about 4° C. for 3 hours. The aqueous and petroleum spirit layers are filtered through a Gooch crucible, and the flask and crucible are washed with petroleum spirit. The remaining precipitate is transferred and washed with

cold water, and the crucible is dried at 100° C. The weight of the benzaldehyde derivative, multiplied by 0.553, gives the corresponding amount of *p*-aminophenol. When recrystallised from alcohol and water, the benzylidene-*p*-aminophenol melts at 185° to 186° C. To separate *o*-aminophenol from *p*-aminophenol and *p*-methylaminophenol, the dry mixture (not exceeding 200 mg.) is dissolved in 2 ml. of glacial acetic acid, 8 ml. of chloroform are added and the mixture is stirred to induce crystallisation and left for 30 minutes. The crystalline precipitate is collected in a Gooch crucible, washed with chloroform, and dried in a desiccator. The weight of *o*-aminophenol acetate, multiplied by 0.645, gives the corresponding amount of *o*-aminophenol. Under the conditions stated, about 2.5 mg. of *o*-aminophenol remain in solution, and this amount may be added as a correction to the quantity found. Experiments with mixtures of known composition showed that the methods yielded results within 5 per cent. of theory. A. O. J.

Colorimetric Estimation of Indigo. J. Lotichius. (*J. Soc. Dyers and Colourists*, 1940, 56, 433-434).—By a modification of Lotichius' method for estimating indigo in indigo vats (*cf. J. Soc. Dyers and Colourists*, 1939, 55, 87), the indigo-content of commercial indigo may be estimated quantitatively as follows:—To a known weight (approx. 0.25 g.) of indigo in a 250-ml. flask, are added 100 ml. of water, 10 ml. of sodium hydroxide solution (sp.gr. 1.425), 7 ml. of sodium protoalbinat solution (*cf. Lotichius, loc. cit.*), and about 2 g. of sodium hydrosulphite powder. After standing stoppered, at a temperature not exceeding 30° C. for some hours (preferably overnight) until reduction is complete, a few ml. of 30 per cent. hydrogen peroxide solution are added. The blue colloidal indigo solution produced is made up to 250 ml. Five ml. are diluted to 1 litre and the colour is matched with that of a standard solution of Sirius Blue B and Sirius Green 2B (*cf. Lotichius, loc. cit.*). Saponifiable organic constituents, which cause turbidity, must first be removed. By this method indigo may also be estimated in indigo printing paste in about 30 minutes. A sample of printing paste was found to contain 4.1 per cent. of indigo, the calculated proportion being 4.2 per cent. E. B. D.

Colorimetric Estimation of Sodium Hydrosulphite and the Hydrosulphite-content of Indigo Vats. J. Lotichius. (*J. Soc. Dyers and Colourists*, 1940, 56, 434-435).—The method is based on that for the colorimetric estimation of indigo (see preceding abstract). To approximately 1 g. of indigo (if in the form of a powder it should be rubbed into a paste with alkaline water) are added a known weight (about 0.5 g.) of sodium hydrosulphite ($\text{Na}_2\text{S}_2\text{O}_4$) and air-free water containing 3 ml. of sodium hydroxide solution (sp.gr. 1.425) to make up 100 ml. After displacement of the remaining air by coal gas the flask is stoppered and shaken for 30 minutes

at room temperature (over-reduction occurs at higher temperatures), and the excess indigo is allowed to settle for a further 5 minutes. Ten ml. of the supernatant liquid are then pipetted into 10 ml. of sodium protoalbinat solution (*cf. Lotichius, loc. cit.*) in another graduated flask, the tip of the pipette being kept below the surface of the protoalbinat solution, and the mixture is oxidised with 1 ml. of 30 per cent. hydrogen peroxide solution, and diluted a hundred-fold, and colorimetrically tested. The factor for calculating indigo into sodium hydrosulphite is 0.66. For the estimation of the hydrosulphite content of indigo-alkali-hydrosulphite vats, the indigo, I_1 (g. per litre) is estimated colorimetrically. The sodium hydrosulphite is then oxidised with excess of indigo, and the reduced indigo I_2 (g. per litre) is estimated colorimetrically as described above, with some modifications in detail. From ($I_2 - I_1$) the amount of sodium hydrosulphite may be calculated. E. B. D.

Estimation of Indigo on the Fibre. J. Lotichius. (*J. Soc. Dyers and Colourists*, 1940, 56, 435).—A piece of cotton, 5 cm. square, dyed in indigo, is treated with 20 ml. of cold conc. sulphuric acid (sp.gr. 1.84) in a porcelain dish for 30 to 45 minutes. In one movement the resulting dark green solution of indigo sulphate is tipped into about 750 ml. of water, and the dish is rinsed in this. The indigo in the colloidal solution, which can be kept for some time, may be estimated after dilution to 1 litre. The standard solution of Sirius dyes (see abstract above) may be used for comparison (although this is not theoretically permissible because absorption coefficients may be different) because experimental results agree with those compared with standard indigo sulphate solution. The latter solution, which, unlike the dye solution, must be freshly prepared for each series of estimations, contains 10 mg. of chemically pure indigo, dissolved (as stated above) in 10 ml. of sulphuric acid and diluted to 1 litre.

E. B. D.

Inorganic

Qualitative Reactions of Salicylaldoxime Derivatives with Inorganic Ions. J. F. Flagg and N. H. Furman. (*Ind. Eng. Chem. Anal. Ed.*, 1940, 12, 520-531).—A comparative study was made of salicylaldoxime and its 5-chloro, 3,5-dibromo, and 5-nitro derivatives in their reactions with 72 inorganic ions in neutral, slightly acid, an ammoniacal solution; the sensitiveness of the tests for copper, nickel, bismuth and lead was determined. It was found that the chloro and nitro compounds do not differ to a marked extent in behaviour from salicylaldoxime; the dibromo compound is unsuitable as reagent, owing to its sparing solubility. Salicylaldoxime and its nitro derivative are equally sensitive in tests for copper, lead and nickel; the chloro compound is a less sensitive

gent. The unsubstituted oxime answers for the detection of bismuth. No definite actions were obtained with the alkalis, alkaline earth metals and most of the metals the third, fourth, fifth, sixth, and eighth groups of the Periodic System. W. R. S.

Determination of Ruthenium with Thionalide. W. J. Rogers, F. E. Beamish and D. S. Russell. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 561-563.)—Ruthenium is quantitatively precipitated from 0.2 to 0.5 *N* hydrochloric acid solution by thionalide (thioglycollic β -aminonaphthalide). The ruthenium solution is heated almost to boiling, treated with excess of an alcoholic solution of the reagent, and boiled until the precipitate coagulates (one atom of ruthenium requires two molecules of thionalide). The precipitate is collected, washed with hot water, charred in a tared porcelain crucible, ignited, reduced in hydrogen in the usual manner, and weighed as metal; no decrepitation or deflagration takes place. Sodium chloride does not interfere, but low results are obtained in solutions containing nitric acid. In the distillation method for the separation of ruthenium from other metals, the ruthenium tetroxide may be conveniently absorbed in 3 per cent. hydrogen peroxide in two ice-cooled receivers placed in series. The distillate is transferred to a beaker, treated with the requisite quantity of hydrochloric acid, boiled until oxygen is no longer evolved, and precipitated with thionalide. The hydrogen peroxide used should be pure and free from acetanilide, otherwise the ruthenium precipitate does not coagulate well and low results are obtained. W. R. S.

Determination of Rhenium in Molybdenite. C. F. Hiskey and V. W. Meloche. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 503-506.)—The finely powdered mineral (4 g.) is attacked with 20 ml. of strong nitric acid in a 250-ml. conical flask, 5 ml. of fuming nitric acid being added after violent action is over. When the attack has subsided, the contents of the flask are heated just below boiling-point and, after red fumes cease to be evolved, 50 ml. of strong hydrochloric acid are cautiously added. The liquid is heated until chlorine is no longer evolved, fresh hydrochloric acid being added as required (125 to 150 ml. in all), and eventually concentrated to not less than 25 ml. The solution is cautiously treated with strong sulphuric acid (75 ml.), and the liquid is transferred to a distilling-flask, and distilled for 2 hours at 260° to 270° C. in a current of steam (2 parts) and carbon dioxide or air (part) until 250 ml. of distillate have been collected in the ice-cooled receiver. The distillate is treated with a slight excess of a potassium bromide solution of bromine to oxidise any sulphur dioxide and, then with 30 ml. of strong hydrochloric acid and thioanate and stannous chloride solutions. ANALYST, 1939, 64, 62). Standards containing 10, 50 and 100% of rhenium are also treated in the same manner. The solutions

are set aside for about 30 minutes, by which time the colour due to a little molybdenum carried over in the distillation will have faded, and the colours are then matched in 100-ml. Nessler tubes. The comparison is repeated after about 20 minutes, as a check on the complete decolorisation of the molybdenum complex. Selenium interferes in the process, selenium dioxide being volatile and reduced by the stannous chloride. It may be removed by precipitation with 1 g. of sodium sulphite from the chloride solution of the mineral prior to the addition of the strong sulphuric acid; when the selenium has settled, the solution is filtered, and the filtrate is concentrated to 25 ml. and treated with 75 ml. of strong sulphuric acid, etc., as described above. The rhenium found by this method in a number of molybdenite samples (chiefly American) ranged from nil to 40 p.p.m. W. R. S.

Determination of Sulphide Sulphur in Presence of other Sulphur Compounds. J. B. Lewis. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 535.)—The determination of sulphide sulphur in solutions containing other sulphur compounds by precipitation as lead sulphide in presence of ammonium acetate (*cf.* Balde-schwiler, *Ind. Eng. Chem., Anal. Ed.*, 1934, 6, 402) gives correct results if the solution is treated with sufficient sodium hydroxide to provide a minimum alkalinity of 2 per cent. This alkalinity is necessary to prevent precipitation of lead sulphite, which is not prevented by the presence of ammonium acetate, even in acetic acid solution. W. R. S.

Amperometric Titration of Fluoride with Thorium Nitrate. A. Langer. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 511-514.)—Minute amounts of fluorine in the form of sodium fluoride can be titrated with 0.01 *N* thorium nitrate solution by the use of the dropping mercury cathode for the graphical determination of the end-point. The solution (50 ml.) should contain up to 0.2 mg. of fluorine and potassium chloride or nitrate (0.1 *M*) as the conducting salt. In smaller volumes of solution, quantities of 0.005 mg. of fluorine may be titrated with 0.001 *N* thorium solution. The error is of the order of one per cent. Lanthanum nitrate may be used instead of the thorium salt, but titration with calcium salt solution is unsatisfactory. W. R. S.

Detection and Elimination of Phosphate in Qualitative Analysis by means of Zirconium Salts. F. K. Pittman. (*Ind. Eng. Chem., Anal. Ed.*, 1940, 12, 514-515.)—A revised process has been based on Curtman's zirconyl chloride method (*Chem. News*, 1924, 129, 299), which depends upon the fact that zirconium (with hafnium) forms a phosphate which is insoluble in strongly acid solution. The filtrate from the hydrogen sulphide precipitation (copper and tin group) is boiled until all hydrogen sulphide has been expelled, and then made up to about 100 ml., cooled to room temperature, neutralised with ammonium hydroxide, and made acid with 5 ml.

of 6 *N* nitric acid. To this acid solution 35 ml. of 0.015 *M* zirconyl chloride solution are added, a few drops at a time, with vigorous stirring throughout the addition. The precipitate is left to settle for 1 minute, decanted on to a very porous filter paper, and washed several times without the aid of suction, with small amounts of a 5 per cent. solution of ammonium nitrate, the washings being added to the filtrate. The precipitate is rejected. The solution will then contain less than 1 mg. of phosphate, and the last traces can be removed by adding 10 ml. of 0.05 *M* zirconyl chloride solution, heating the mixture just below its boiling point for 2 minutes, leaving it for 5 minutes and filtering. The filtrate is then treated as if phosphate had not been present. The small excess of zirconium will follow the iron and will not interfere with the usual ferrocyanide or thiocyanate tests for that metal.

In testing for phosphate a small portion of the filtrate from the copper and tin group is treated as directed for the removal of phosphate, except that the zirconyl chloride solution should be 0.005 *M* rather than 0.015 *M*. If very small amounts of phosphate are to be tested for, the main portion of the solution should be evaporated to about 15 ml. before the test portion is taken. In this way less than 1 mg. of phosphate can readily be detected. Iron, chromium, manganese, cobalt and nickel can readily be detected in the filtrate from the phosphate, if present to the amount of 1 mg. in the original solution; for aluminium and zinc, 2 and 5 mg., respectively, are required. There is no serious loss of alkaline earth metals. Except with arsenate, oxalate and ferrocyanide, no precipitate is formed when zirconyl chloride is added to a solution of any of the other common anions. Arsenate gives a white precipitate and ferrocyanide a yellow precipitate. Oxalate gives a transient white precipitate in weakly acid solutions.

Adsorption of Malachite Green by Clays and Allied Materials. V. L. Bosazza. (*Nature*, 1940, 146, 334.)—Certain minerals may be identified by the colours of the adsorption complexes produced by staining with a 0.05 per cent. solution of malachite green for 10 minutes and drying. Examples are:—kaolinite, deep blue-green; anauxite, light to deep blue-green; montmorillonite and beidelite, marine blue; pyrophyllite and talc, blue-green; margarite, no pronounced colour; quartz (which is commonly associated with these minerals), no colour. Pleochroism is pronounced in kaolinite, less marked but variable with anauxite, and does not occur in the other minerals mentioned. Washing with water, acid or alkali fails to remove the colour from kaolinite, although acid changes it to bright red. The extent of the adsorption varies according to the mineral concerned, and in some instances (e.g. anauxite) with its composition. Thus, under the above conditions, white kaolin (rich in kaolinite) adsorbs 85 per cent. of the dye in 1 minute and 93 per cent.

in periods of 60 minutes and over, whilst bentonitic clays and fuller's earth adsorb the dye from much stronger solutions. J. G.

Physical Methods, Apparatus, etc.

Cross Sections of Pulp Fibres. J. H. Graff, M. A. Schlosser and E. K. Nihlen. (*Paper Trade J.*, 1940, 111, T.A.P.P.I. Sect., 105-110.)—The possibility of identifying fibres used in paper manufacture from certain characteristics of their cross-sections was investigated. A small bundle of the fibres in question is desiccated and mounted in paraffin wax; and the cross-sections (diameter 5 microns) are cut and magnified (100 diameters), and drawings of them are made. The average "roundness factor" is then given (as a percentage) by $100 \times \text{gross area of each fibre} / \text{area of a circle whose diameter is the width of the fibre}$; the larger this factor the "rounder" is the fibre. The gross areas of the fibres and the areas of the lumens were determined by means of a planimeter. Other data which may be calculated are the average number of fibres per unit area, area of the lumen and total area of air-space between the fibres when these are assumed to be "stacked" in a pile in the same direction so that their cross-sections are all in the same plane. Results for 10 different types of pulp fibres are tabulated, and statistical treatment of them shows that the values obtained are typical of the fibre concerned, and may be used as an aid to identification. Good agreement was obtained in duplicate determinations. Since the fibre-width is correlated with at least 6 of the other factors, it may prove possible to use it to calculate some of these, but more measurements are required before the conversion factors can be ascertained. The differences between the above values may also provide useful information about the treatment undergone by the fibre during manufacture into paper. J. G.

Accelerated Spray Test for Determining the Relative Atmospheric Corrodibility of Ferrous Materials. T. Swinden and W. W. Stevenson. (*Advance Copy Paper*, No. 2, 1940, of the Corrosion Committee, Iron and Steel Inst., 1940.)—A method of automatic accelerated spray testing is described, in which the specimens, on an endless belt, complete a cycle of treatment, consisting in spraying, followed by drying in air and in a warm chamber. The cycle is completed in 7 minutes, and the specimens are subjected to 60 cycles per day. A standardised douching with water is given at the beginning of the second and subsequent days, and the daily spraying and drying treatment is continued for 20 days. Comparative results of long-period field tests and spray tests are given for 14 irons and steels of the low-alloy constructional type. Good agreement is shown in the orders of merit obtained in one-year field tests and in the 20-day spray tests, using a spray liquid consisting of *N*/100.

sulphuric acid and *N*/100 sodium chloride. Less satisfactory agreement is shown in field tests and spray tests, using *N*/100 and *N*/20 sulphuric acid without sodium chloride. It is suggested that the results obtained with the

sulphuric acid and sodium chloride solution are of sufficient promise to encourage other workers to re-examine the spray method as the basis of a satisfactory short-time laboratory corrosion test.

Reviews

CONVERSION OF PETROLEUM. By A. N. SACHANEN, D.Sc. Pp. 413. New York: The Reinhold Publishing Corpn.; London: Chapman & Hall, Ltd. Price 36s. net.

The subject of this book, as the name suggests and as set out in the sub-title, is "Production of motor fuels by thermal and catalytic processes," that is, by cracking and recently developed methods such as hydrogenation and isomerisation with the aid of catalysts.

Dr. Sachanen is a member of the research and development division of the Socony Vacuum Oil Company, and is, as the book bears out, well qualified to write fully and authoritatively on petroleum conversion. The subject being so large, the author has wisely confined himself to the essentials, and has omitted reference to products, processes and plant and equipment generally used in other sections of the petroleum industry.

The book is divided into seven chapters, of which the first three, taking up more than half the book, discuss the fundamental factors and the thermal and catalytic reactions on which the design of plant and its operation depend. The subsequent chapters deal with the plant and equipment used and with the properties and chemical treatment of the new types of motor fuels produced, followed by a concise chapter on cracked products other than gasoline, which, although relatively of little use to-day, will in the future undoubtedly prove to be suitable for special purposes.

The book opens with an account of the paraffins, and the chapter gives an outline of the reactions of dehydrogenation, splitting decompositions, catalytic cracking, the so-called alkylation, isomerisation, and aromatic cyclisation. Data obtained by the many research workers in these fields are supplemented by those from actual operating plants. The olefines are treated in a similar way, polymerisation with and without catalysts, hydrogenation, isomerisation, and cyclisation being discussed. A description of the commercial methods in these processes is also given. The naphthenes, defined as "all saturated cyclic hydrocarbons, monocyclic and polycyclic, which have not the aromatic character," are discussed in relation to dealkylation, alkylation, dehydrogenation, ring rupture and isomerisation. Aromatic hydrocarbons are similarly treated, the reactions of a number of hydrocarbons particularly characteristic of coal-tar being included. Short summaries are given of each of these sections, which are very helpful. Only brief reference is made to the oxygen, sulphur and nitrogen compounds, these being at present of relatively little importance and interest; there is a very full bibliography at the end of this important chapter. The references are mainly to work carried out during the last ten years.

In the second chapter the author discusses the various factors that influence the progress of the reactions, the effects of temperature and pressure on the main and secondary reactions, the formation of coke and the effects of recycling. Residue- and non-residue-cracking, viscosity breaking, and reforming are also dealt with, the data quoted in support being critically examined.

Hydrogenation forms the subject of a separate chapter which covers the whole subject completely, including applications of the process, such as the conversion of low-grade lubricating stocks and Diesel oils into high-grade products,

as well as the conversion of heavy asphaltic crudes and refinery residues into gasoline and gas oils. Much information is given on the subject of the catalysts used, but little concerning the actual plant, few details being as yet available for publication.

The equipment used for the cracking operations is well described and illustrated by reference to well-known types of plant in successful operation. Processes which have been of importance in the early development of cracking are not described, being mainly of historical value.

The development of cracking has given rise to problems of chemical refining much more complex than those which obtained in the days of only straight-run products; they take up two full chapters. The determination of the relative proportions of the various types of hydrocarbons in a complex mixture presents a problem that is at present far from being solved; data given exemplify this. It is only to specially fractionated cuts of very narrow boiling-point range that certain methods can be successfully applied. The complex questions of gum formation, inhibitors, octane numbers and susceptibility to lead tetra-ethyl are adequately discussed.

The book contains a mass of detail well selected, set out, summarised and often criticised. It is valuable, therefore, not only as a work of reference, but as one that can be read with pleasure. It should prove of great use, not only to the average petroleum chemist and works operator, but also to those who have specialised in one or more of the many aspects of this great modern industry.

J. KEWLEY.

TEMPLES OF ARMANT. By SIR ROBERT MOND, LL.D., D.C.L., F.R.S., and OLIVER H. MYERS. 2 vols. Pp. xii + 223: 107 plates. London: The Egypt Exploration Society. 1940. Price £3 3s.

Two reports on Armant by the same authors have already been published, namely, "*The Bucheum*" and "*Cemeteries of Armant I*" (cf. ANALYST, 1935, 60, 65; 1938, 63, 690), Sir Robert Mond financing the work and Mr. Myers, with the help of various assistants, undertaking it.

Armant is a town in Upper Egypt, about twelve miles south of Luxor. Anciently it was of considerable importance, though now it is only a small place, noted chiefly for its sugar factory. During the Christian period in Egypt Armant was the seat of a bishopric, and the church (of which only a few ruined columns remain) was one of the largest and finest in Egypt.

The book consists of a preface and twelve chapters, in the writing of which the chief author has had the help of five collaborators. Mr. Myers regrets that there are fewer analyses and expert reports than in his previous books, though these are by no means lacking, and the last chapter consists of technical reports by sixteen different experts, who respectively deal with pottery; glass; glass-making materials; metals; metal-smelting products; geology; mineralogy; zoology; wood; osteology. There is also a report, by Mr. Myers himself, on weights.

As the reviewer is a chemist and the review is for THE ANALYST, the chemical and allied aspects of the report will be especially considered. A description is given of the methods employed for cleaning and preserving carved and painted limestone (both *in situ* and in the laboratory), and also of the manner in which a bronze lamp, too fragile for ordinary methods, was dealt with, and Mr. Harden recounts how he treated decayed glass mosaics.

Mr. Myers finds a difficulty in explaining the fact that certain pottery, although overfired and cracked in the kiln, was still black in the centre, like ordinary under-fired ware. I would suggest that probably there was a large proportion of alkali present, which became fused round the charred organic matter and so prevented its complete combustion.

A green corrosion product on a silver-copper alloy is called by one expert "verdigris," which he defines as a "mixture of basic copper carbonate and acetate," but that there was any copper acetate in this particular instance was not proved and seems unlikely, the expert apparently having been misled by the frequent, but wrong, use of the word "verdigris" to describe the similar-looking basic copper carbonate, or sometimes even copper oxychloride.

No book is, or can be, perfect, and a few minor points may be criticised. Thus, the German word *Fundplatz* occurs more than fifty times, and one of the experts uses its English equivalent "find-place," although the words "place"; "position"; "situation" and "locality" are available. The use of the French word *sondage* in place of "sounding" or "boring" is equally unnecessary and objectionable. Several Arabic words, for example, *gisr*, *maktub* and *saqqia*, are used without any explanation of their meaning. Also, why employ the word *insulae*? On p. 23 the text reads "to let the plaster," a word or words apparently having inadvertently been dropped. Mr. Myers has invented a new verb, namely, "to corpus" with "corpusing," which is both unnecessary and ugly. Also he says that certain metal objects were "dissolved" by the treatment used, whereas "disintegrated" is meant.

These criticisms, however, are of small matters, and the chief author is to be congratulated on an excellent report that compels admiration for the detail with which the work was done and the manner in which it is recorded, for the orderly arrangement of the vast amount of material and for the way in which, by submitting what often must have seemed most unpromising specimens to experts, he has caused to be extracted from them useful and profitable results. It is manifest that an immense amount of hard work, which would have daunted many archaeologists, has been done both in the field and in the preparation of the report. The historical summary is interesting as well as valuable.

The book is well printed and bound, and three of the plates are in colour. The inclusion of a plate showing sixteen blocks of carved limestone from Armant, now in the Grenoble Museum, is a happy inspiration.

A. LUCAS.

A TEXT BOOK OF BACTERIOLOGY. By R. W. FAIRBROTHER, D.Sc., M.D. 3rd Edition. Pp. 451 + x, 6 plates. London: William Heineman (Medical Books), Ltd. 1940. Price 17s. 6d.

This book is in the main an outline of the medical aspects of bacteriology. In writing it the author's aim has been to restrict himself to branches of bacteriology of medical importance and to the application of bacteriological methods to the prevention, diagnosis and treatment of disease, while avoiding excessive technical detail, which, although required in the laboratory, is not necessary for the average medical man. There is, however, a concise account of general bacteriological technique and of general bacteriology, amply sufficient in detail for all students of bacteriology. The book is divided into three parts under the following headings: —I General Bacteriology, II Systematic Bacteriology, and III General Technique.

Part I contains twelve chapters, beginning with a historical survey and then discussing the biology, morphology, cultivation, multiplication and destruction of bacteria, and their classification. Other chapters deal with antigens, hypersensitiveness, idiosyncrasy, and allergy, and the relation of bacteriology to medicine. In Part II (22 chapters) there is a systematic description of the different pathogenic organisms, concluding with chapters on filterable viruses, the bacteriophage and the bacteria of water, milk and shell fish. Part III comprises three chapters, dealing respectively with the microscope and staining methods, the preparation of culture media, and serological technique.

The chapters in Part II are subdivided into sections and paragraphs dealing with the classification of the organism, its habitat, morphology and

staining reactions, cultural reactions, resistance, biochemical activity, serology, toxin production, pathogenicity, diagnosis, specific therapy and chemotherapy. This is often followed by a concise account of the disease or diseases which the organism causes, with bacteriological diagnosis, prophylaxis and therapy. In this way, under one or other of the headings, all the important points concerning a micro-organism and its related disease are dealt with, if only briefly.

The book is well up-to-date; for example, one finds reference to the relationship between virulence of strains of *Staphylococci* and production of coagulase, and to the modern typing of meningococci and Flexner dysentery bacilli. There is also a good description of Wilson and Blair's medium for the isolation of *B. typhosum*. Obviously a book of this size cannot contain everything, but a description of the valuable method of E. R. Jones, using tetrathionate broth and brilliant-green eosin agar, would be well worth including.

The book is very well written and arranged, the subject matter is well balanced and clearly expressed, the printing is in good bold type, and the paper and binding are excellent. The volume can be strongly recommended; it has been justly described by other reviewers as the best of its kind.

D. R. WOOD

CALCULATIONS OF QUANTITATIVE ANALYSIS. By CARL ENGELDER, Ph.D. New York: John Wiley & Sons, Inc.; London: Chapman & Hall, Ltd. 1931. Pp. viii + 174. Price 12s.

This book opens with a discussion of general matters such as errors, precision and accuracy. It deals very fully with the use of logarithms, numerous worked examples being given. Reagents and methods of expressing the strength of a solution also receive attention. Considerable stress is laid on the laws of mass action and combining weights. Part 2, under the headings of Precipitation, Neutralisation and Oxidation, is devoted to the calculations involved in volumetric analysis. In Part 3 the theory of gravimetric analysis is discussed, and the last section is devoted to the calculations required in systematic quantitative analysis. Numerous useful tables including logarithms, solubilities, densities, and ionisation constants, are given. A satisfactory index is also given.

Each section contains a very clear treatment of the appropriate underlying theory. Thus in the section on neutralisation we find a very full discussion of ionisation, pH value and titration curves. Oxidation-reduction processes are treated from the point of view of electron transfer, and in gravimetric analysis the concept of solubility-product is used. Attached to each section is a set of excellent numerical examples, making in all a total of three hundred.

Since this book is intended as a theoretical adjunct to systematic laboratory work, no practical directions are given. Used as suggested by the author, it will greatly enhance the value of practical work and convert analytical procedure, which is too often regarded as a mystic ritual, into an intelligent occupation. The treatment is modern and has therefore a definite physical bias. It is safe to predict that one who has faithfully worked through the suggested course will have acquired a satisfactory foundation for advanced work.

HAROLD TOMS

